

**U.S. Fish and Wildlife Service  
Region 3  
Division of Environmental Contaminants**

**CHEMICAL, BIOLOGICAL AND  
TOXICOLOGICAL INVESTIGATION OF  
OIL DRILLING IMPACTS TO  
AQUATIC RESOURCES  
IN THE YELLOWBANK SLOUGH REGION  
NEAR NEW HAVEN, ILLINOIS**

**Melanie Y. Young**

**Study Identification Number 90-3-104**



**U.S. Department of the Interior  
Fish and Wildlife Service  
4469 48th Avenue Court  
Rock Island, Illinois 61201**



**February 1993**

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### Abstract

A survey of contaminants in sediments and aquatic macroinvertebrates at several locations on the proposed Yellowbank Slough National Wildlife Refuge was conducted during the 1990 and 1991 field seasons. Surface water samples, bottom sediments and aquatic macroinvertebrates were collected at selected sampling locations for chemical analysis and aquatic invertebrate bioassays. This paper reports the concentrations of inorganic and organic chemical parameters in bed sediments, the results of Ceriodaphnia dubia and Hyalella azteca bioassays, and the results of the Microtox™ assays for sediment and water samples collected from selected locations in the drainage. Although widespread contamination due to oil and gas development activities was not apparent from the data obtained during the present study, the results indicate that there may be some potential localized areas of concern throughout the proposed area of acquisition in the Yellowbank Slough region.



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## Introduction

Yellowbank Slough is an extensive region of emergent wetlands and bottomland forest located in southeastern Illinois at the juncture of the Ohio and Wabash River watersheds. It has historically provided migration and wintering habitat for a significant portion of the Ohio River Valley waterfowl migration during the fall, winter and spring when floodwaters from the Wabash River inundate the region. Yellowbank Slough is currently designated as a high priority North American Waterfowl Management Plan Lower Mississippi Region Joint Venture project site [Illinois Dept. of Conservation 1988; U.S. Fish and Wildlife Service 1986(a); U.S. Fish and Wildlife Service 1986(b)]. Yellowbank Slough is a proposed National Wildlife Refuge (NWR) and is located in a heavy crude oil and gas production region of southeastern Illinois near New Haven. A biological, chemical and toxicological survey was conducted in 1990 and 1991 to assess potential impacts to aquatic resources resulting from exposure to petroleum hydrocarbons and associated wastes generated from surrounding previously active and currently producing oil fields. This study was designed to identify potential regions of environmental contamination originating from several critical crude oil point sources within the primary Yellowbank Slough drainage. Baseline data was compiled on organic and inorganic contaminants in bottom sediments and aquatic invertebrate tissue, and the acute and chronic toxicological potential of surface water and bed sediment samples collected from selected sites within the region.

Illinois has over 400 active and abandoned oil fields located throughout the southern half of the state. Historically, oil and gas development has been conducted in Illinois since the early 1900s. The oil and gas fields are located primarily in the southeastern portion of the state (Figure 1) and extend geographically over approximately 42 counties. By far the most significant oil and gas production areas are currently located in White, Lawrence and Wayne counties. Yellowbank Slough is located at the southernmost edge of a larger oil production region in Gallatin County, Illinois.

Sources of contaminants to surface waters in this region include waste brine fluids and crude oil from normal operations and accidental spills; major accidental resurfacing of oil and brines during secondary and tertiary recovery operations; and drilling fluids temporarily and permanently stored in on-site and off-site surface impoundments. Anticipated organic contaminants resulting from these activities and operations are



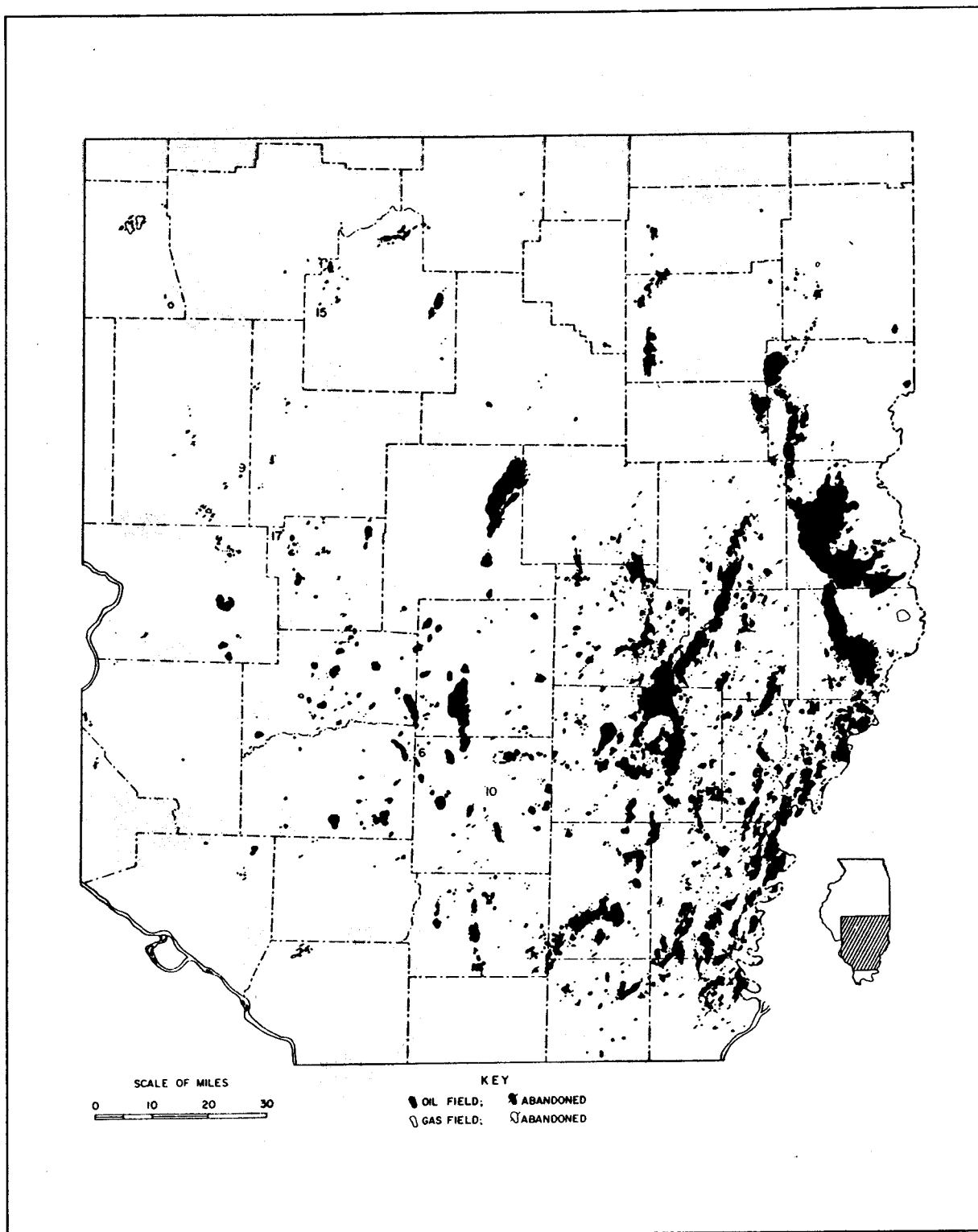


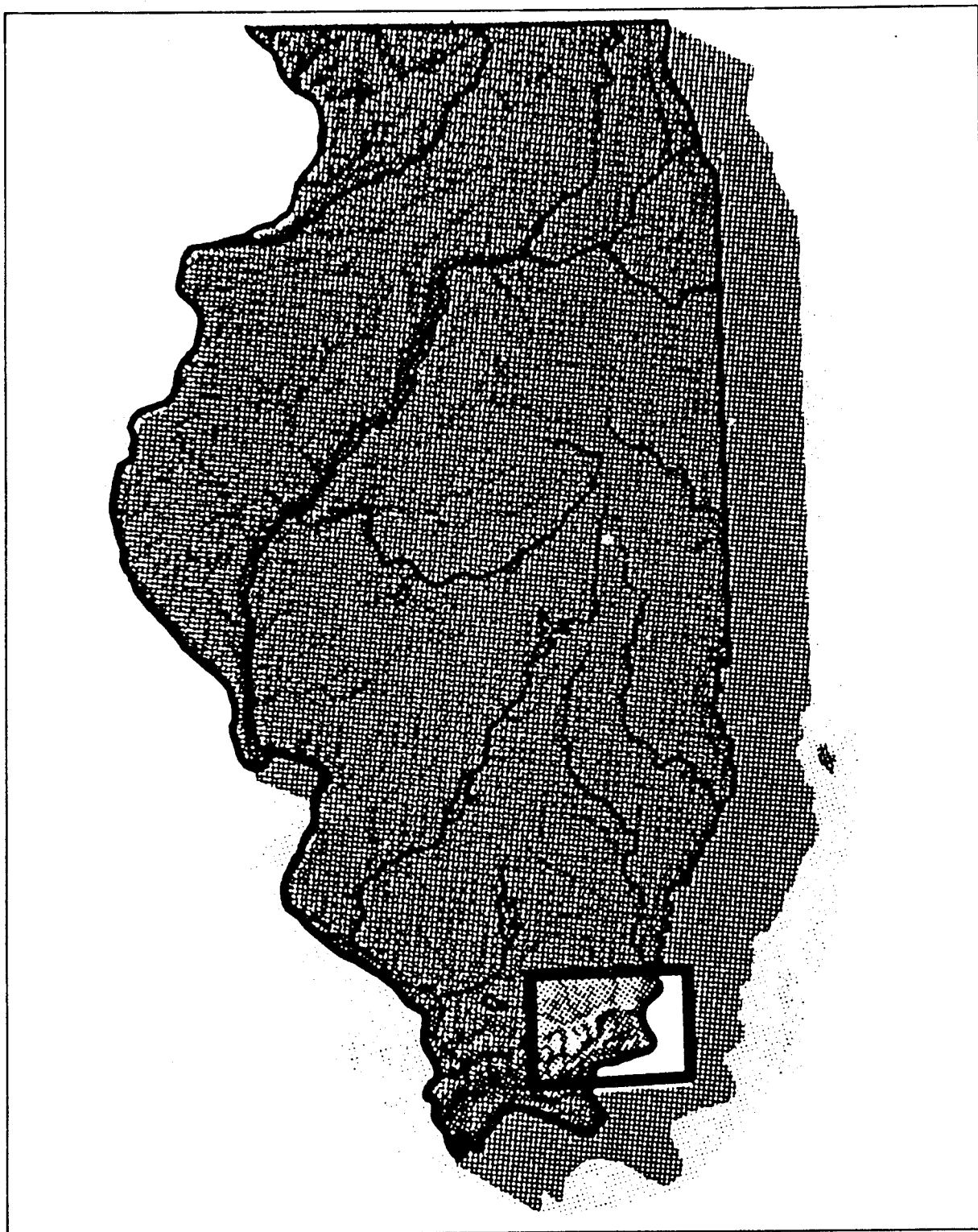
Figure 1. Oil and gas production region of Illinois.

those chemicals derived from petroleum, principally aliphatic and aromatic hydrocarbons, including polycyclic aromatic hydrocarbons (PAHs). Crude oil is generally composed of saturated hydrocarbons (including straight and branched alkanes and cycloalkanes), aromatic hydrocarbons (including polycyclic aromatics, cycloalkanoaromatics and cyclic sulphur compounds), and resins and asphaltenes (including high molecular weight polycyclic fractions containing nitrogen, sulphur and oxygen compounds) (Gill and Robotham 1989). Although composed predominantly of saturated hydrocarbons, some crude oils may contain as much as 25% (by weight) aromatic hydrocarbons. Geologic materials in the general area can be described as alluvium, composed of a mixture of highly permeable sand, gravel, silt and clay deposits, with a significant potential for surface water contamination of shallow aquifers. Petroleum-based hydrocarbons that were historically discharged directly and/or via groundwater to surface waters might be anticipated to partition in particulate matter, and, to a lesser extent, water. Generally the alkanes are less water soluble than other components of crude oil; the aromatic hydrocarbons form the primary component in water soluble fractions, with low-molecular weight compounds exhibiting greater solubility than higher molecular weight compounds (Robotham and Gill 1989). These hydrocarbons may be available for uptake by some benthic aquatic organisms. PAHs are known to adversely affect some fish species in metabolized forms, and have been previously demonstrated to be acutely toxic to various macroinvertebrates (Crunkilton and Duchrow 1990; Woodward et al. 1987). They can accumulate to relatively high proportions in lower trophic level organisms unable to metabolize them, such as some aquatic invertebrates (Eisler 1987).

Oil and gas exploration per se may not necessarily have a major adverse effect on fisheries and wildlife resources. Rather, it is the indirect or residual effects of oil drilling that could create potential hazards for biota in any given region (Woodward et al. 1988), and in the Yellowbank Slough region in particular. Crude oil has previously been demonstrated to be toxic to wildlife, as evidenced by catastrophic fisheries and waterfowl mortalities observed during and after major oil spills (Piatt et al. 1990). Although oil drilling is regulated in the State of Illinois, contamination of terrestrial and aquatic habitats can occur through many routes: pumping accidents, local transportation accidents, storage tank breaches and broken pipelines, to identify a few. Of these, storage tank leaks and suspected broken pipelines were observed to have occurred in the Yellowbank Slough region during the present study.

## Study Area Description

The Yellowbank Slough region is situated within an approximately 20,000 acre tract located near the confluence of the Wabash and Ohio Rivers in Gallatin County, Illinois (Figure 2). The regional drainages are generally located in an area of oil and gas development that contains extensive abandoned and currently active oil wells, in addition to natural gas and other service wells (Figure 3) (Illinois State Geological Survey 1986). The Yellowbank Slough study area in which the field investigation was conducted consists of the main stream channels through Yellowbank Slough and Running Slough. An off-site reference site was selected in the same general vicinity of the study area located near Shawneetown, Illinois that did not show any visual evidence of contaminants present due to current or past oil drilling activities. The sampling locations for the present study are shown in Figure 4 and listed in Table 1. Eleven sampling locations were selected within the study area which represented a variety of habitat types within the Yellowbank Slough and Running Slough drainages. An upstream reference sampling location, site YS-1, and an off-site reference location, site YS-12, were selected and sampled in order to better compare and contrast data obtained from various sample sites within the drainage. Sections within the upper end of the slough, the mid-section of the slough and the lower end of the slough were each sampled. Additionally, some areas adjacent to crude oil storage tanks and actively pumping wells encountered during field reconnaissance showed evidence of recent or on-going crude oil discharges to the aquatic environment. These areas were also sampled to assess the degree of adverse impact to adjacent aquatic resources.



**Figure 2. Yellowbank Slough region in southeastern Illinois.**

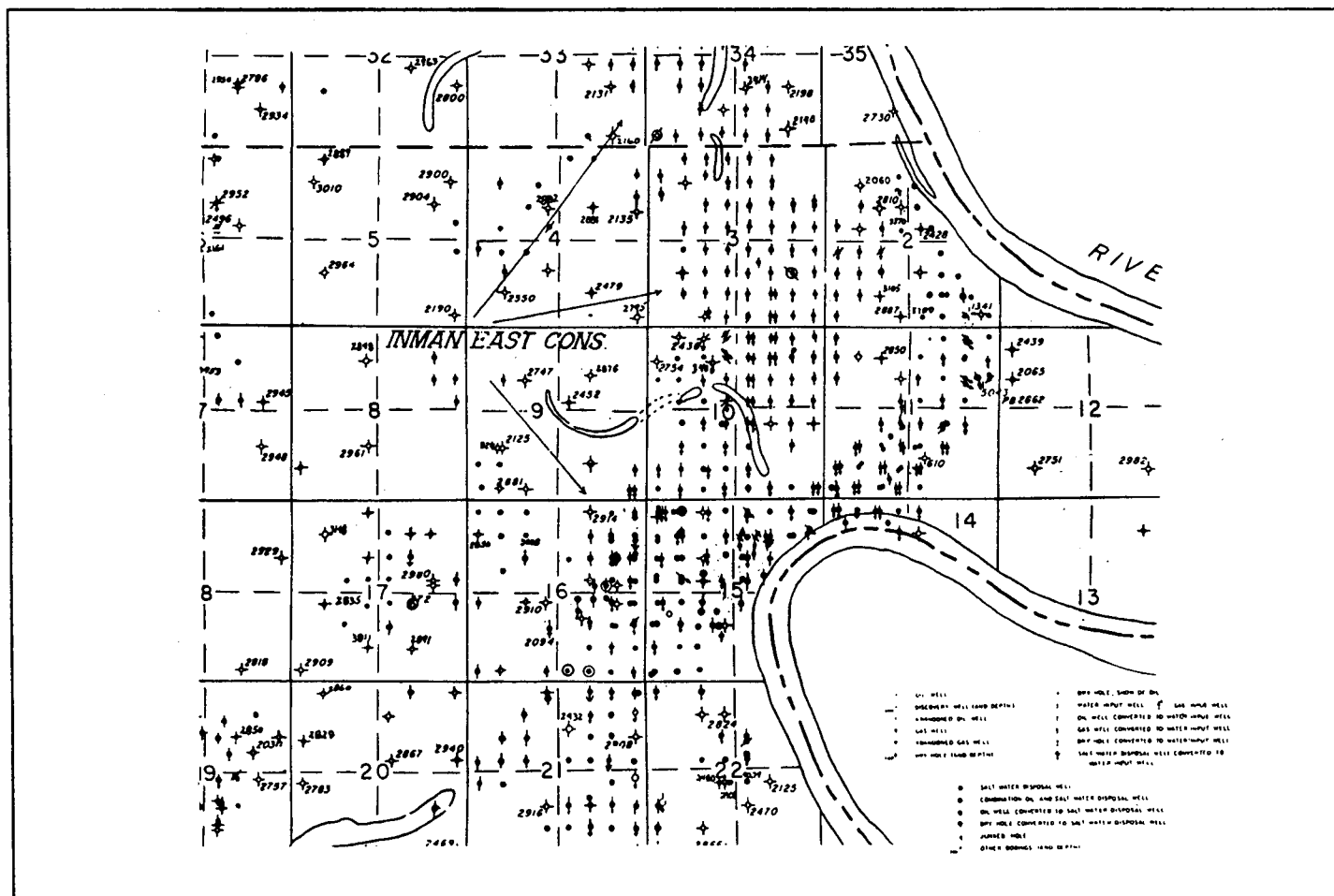


Figure 3. Oil wells in the Yellowbank Slough region (Illinois State Geological Survey 1986).

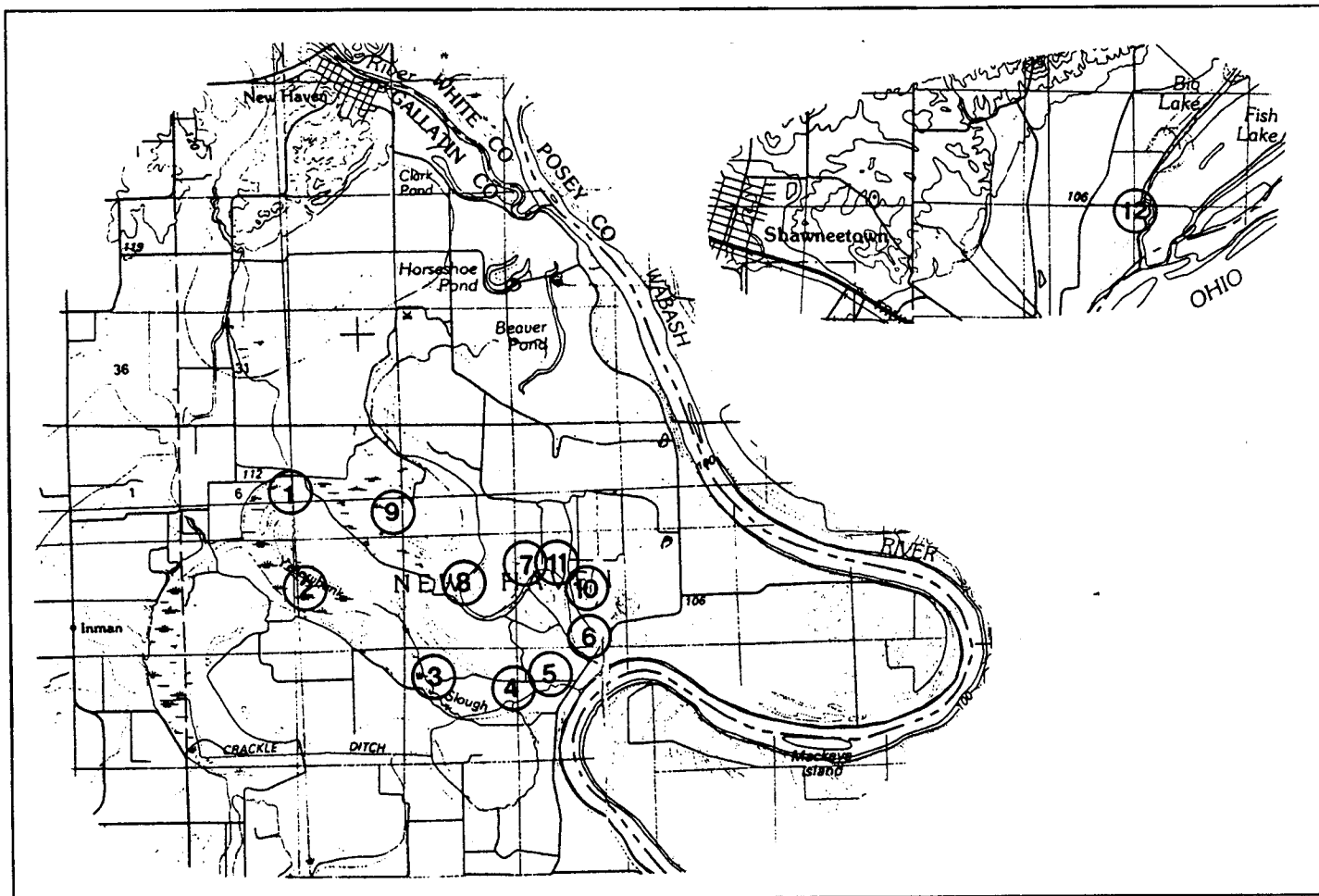


Figure 4. Yellowbank Slough study area sampling sites (sites are numbered 1 - 12).

Table 1. Yellowbank Slough region 1990-91 survey sampling sites.

Sample Location	Sample Number	Sample Type(s) Collected	Toxicity Tests Conducted
Upper NW end of Yellowbank Slough (SE 1/4, Sec. 6) (upstream reference)	YS-1	Bottom sediments Benthic macroinvertebrates  Artificial substrates	<u>Ceriodaphnia dubia</u> (surface water) <u>Hyallolella azteca</u> (sediments) Microtox (sediments)
Upper NW end of Yellowbank Slough (W 1/2, Sec. 8)	YS-2	Bottom sediments	
Middle section of Yellowbank Slough (NW 1/4, Sec. 16)	YS-3	Bottom sediments	
Lower section of Yellowbank Slough (W 1/2, Sec. 15)	YS-4	Bottom sediments	<u>Ceriodaphnia dubia</u> (surface water) <u>Hyallolella azteca</u> (sediments) Microtox (sediments)
Lower section of Yellowbank Slough (Central Sec. 15)	YS-5	Bottom sediments	
Lower section of Running Slough	YS-6	Bottom sediments	
Middle section of Running Slough	YS-7	Bottom sediments Benthic macro-invertebrates Artificial substrates	<u>Ceriodaphnia dubia</u> (surface water) <u>Hyallolella azteca</u> (sediments) Microtox (sediments)
Middle section of Running Slough	YS-8	Bottom sediments	
Goose Pond	YS-9	Bottom sediments Crayfish	<u>Ceriodaphnia dubia</u> (surface water) <u>Hyallolella azteca</u> (sediments) Microtox (sediments)
Lower section of Running Slough	YS-10	Bottom sediments	
Crude oil spill site (positive control)	YS-11	Bottom sediments	
Shawneetown vicinity drainage (negative control)	YS-12	Bottom sediments Benthic macro-invertebrates Crayfish	<u>Ceriodaphnia dubia</u> (surface water) <u>Hyallolella azteca</u> (sediments) Microtox (sediments)

## Methodology

### Sample Collections

Bottom sediment samples were collected for bulk chemical analysis during the 1990 field season at 11 designated sampling locations in the Yellowbank Slough drainage (Table 1). One composite bottom sediment sample, comprised of three to five subsamples, was collected at each site. Sediments were collected with an Ekman bottom dredge sampler with 6" x 6" x 6" chamber dimensions. At a few of the sites where extremely shallow water conditions prevailed, sediment samples were collected by hand with a stainless steel spoon and stainless steel pans. The subsamples for each site were placed in previously acetone-rinsed stainless steel containers and thoroughly mixed, then proportioned into 1000 ml chemically cleaned borosilicate glass jars with Teflon™-lined lids. The sampling equipment was rinsed with acetone between each sample site location. The sample containers were labeled and temporarily stored at ambient temperature in the field during sample collections, prior to being transported back to the laboratory and stored at -20° C. The samples were then later shipped frozen under dry ice to the analytical facility for chemical analysis.

Aquatic macroinvertebrates were collected for chemical analysis during the 1990 field season at one site within the study area, one reference site outside of the study area, and one upstream study area reference site. A diverse mixture of benthic invertebrates consisting primarily of Physa spp., Odonata larvae, and Corbicula spp., was collected at the upstream reference site (YS-1), and at the off-site reference location at Shawneetown (YS-12). Crayfish (Procambarus spp.), which represent higher trophic level predatory invertebrates, were collected at one sampling location in the study area (YS-9), and at the off-site reference location (YS-12). The crayfish were collected with modified minnow traps baited with chicken wings.

Approximately 8-10 specimens were placed in a single chemically cleaned glass jar which comprised one sample for each site. The benthic invertebrates collected at each site were identified to sub-order, family or genus level and composited to make up a sample of at least 500 grams for chemical analysis. The invertebrates were collected with an Ekman bottom dredge sampler, and the samples were sieved through a WILDCO™ benthos wash bucket until a sufficient quantity was obtained for



chemical analysis. The sieved invertebrates were composited into 1000 ml chemically cleaned borosilicate glass jars with Teflon-lined lids which were labeled by sampling location. The sample containers were temporarily stored at ambient temperatures in the field, transported back to the field office, and stored at -20° C prior to shipment to the analytical facility.

Because of the importance of macroinvertebrate diversity and density in an aquatic community to waterfowl, particularly breeding waterfowl (Eldridge 1990), and because changes in macroinvertebrate community structure may be indicative of contaminant-related stress (U.S. Environmental Protection Agency 1990), benthic aquatic macroinvertebrate diversity in Yellowbank Slough was sampled during the 1990 field season at two sites using two bottom sampling methods: artificial substrates and bottom dredge samples of benthic invertebrates. Hester-Dendy artificial substrates were placed at two sampling sites in the study area which represented physically similar habitat types: at the upstream reference site YS-1, and at approximately the mid-section of Running Slough at site YS-7. Each of the plates in a unit were 3" x 3" in surface area, each unit providing a total surface area of approximately one square foot. Two substrates were suspended from a float positioned approximately halfway between the water surface and the stream bed at each site for a total of four weeks. At the end of the four-week colonization period, the substrates were collected and placed in 1000 ml glass jars in 10% buffered neutral formalin in the field, and later transferred to 70% ethyl alcohol in the laboratory. Benthic invertebrates were also sampled at the upstream reference site YS-1, and approximately at the mid-section of Running Slough at site YS-7. Benthos were sampled using an Ekman bottom dredge sampler with 6" x 6" x 6" chamber dimensions. Five sub-samples were collected at each site. The subsamples were sieved with a WILDCO benthos wash bucket to remove excess debris, and composited into two or three labeled 1 gallon borosilicate glass jars per site. The benthic invertebrate sample jars were then filled with 10% buffered neutral formalin in the field, and later transferred to 70% ethyl alcohol after being transported back to the laboratory. The preserved invertebrates from both sets of samples, the artificial substrates and the benthos, were then transferred to the Iowa Hygienics Laboratory in Des Moines, Iowa where they were enumerated and identified to the family taxonomic level by personnel highly experienced in invertebrate taxonomic identification.

Surface water samples and bed sediments were collected for toxicity tests and bioassay analysis during the 1991 field season. Water samples and bed sediments were collected at four sampling sites in the Yellowbank Slough region, and at the reference site in the Shawneetown area. Surface water samples were collected for 96-hour bioassays using the cladoceran, Ceriodaphnia dubia. Bed sediments were collected for 10-day bulk sediment toxicity tests with the epibenthic amphipod, Hyalella azteca, and bioassays using the Microtox™ Solid-Phase and 100% Tests Protocols (Microbics Corp. 1991), which employ the luminescent marine bacterium Photobacterium phosphorium. The sediment samples were collected by mixing and compositing subsamples in stainless steel containers rinsed successively in distilled water, hexane and 5% nitric acid, and then proportioning those composites into 1000 ml chemically cleaned borosilcate glass jars with Teflon-lined lids. Each sample was labeled and stored chilled temporarily in the field. Upon completion of the collections, the samples were immediately shipped chilled to the toxicity testing facility and stored at 4° C until preparation for the bioassays. Pore water extracts for the bioassays were prepared according to the methods decribed by Giesy et al. (1988). All sediment exposure bioassays using H. azteca were initiated within two weeks of initial sample collections. Water samples collected for the 96-hour C. dubia bioassays were collected and stored in one liter Nalgene™ containers, shipped immediately to the toxicity testing facility, and stored at 4° C until the bioassays were started the following day. The sediment samples collected for the Microtox™ tests were kept chilled in the field, then immediately transferred to the field office and stored at 4° C until preparation for the bioassays, which were completed within two weeks of initial sample collections.

General surface water quality measurements were taken in the field at various locations in the Yellowbank Slough region during the 1991 field sampling. Water quality characteristics measured were temperature (°C), pH, dissolved oxygen (ppm), specific conductance (umhos), and salinity. Temperature and dissolved oxygen were measured with a YSI Model 51B oxygen meter. The pH was measured with a Hach™ Pocket Pal pH tester (catalogue number 44350). Specific conductance and salinity were measured with a YSI Model 33 S-C-T meter. All meters were calibrated to manufacturers' specifications prior to use.

## Chemical Analysis

Bed sediment and aquatic invertebrate samples were analyzed for trace elements, aliphatic hydrocarbons, polycyclic aromatic hydrocarbons, organochlorine pesticides and polychlorinated biphenyls (PCBs).

Bed sediments were chemically and physically characterized by percent moisture, total organic carbon (TOC) content, and grain size analysis. Determinations were made by Environmental Trace Substances Research Center, Columbia, Missouri. Percent moisture determinations were made by oven-drying a pre-weighed homogenized aliquot of the sample in a Fisher Isotemp oven at 103-105° C, and then reweighing the dried sample. Total organic carbon analysis was made using a dry oxidation method. The samples were treated with 10% HCL to remove the inorganic carbon prior to TOC analysis, with concurrent blank preparation. The samples were oven-dried at 100° C for at least one hour, and then analyzed using a standard curve prepared with a set of potassium hydrogen phthalate standards, which also received the same preparation as the samples. The micrograms of organic carbon determinations in each sample were made using this potassium hydrogen phthalate standard curve. Results were reported by the analytical facility in units of percent concentration. The limits of detection for a 4 mg sample was 0.1%. The accuracy and precision of the analysis was measured by method blank sample analysis, spike recovery analysis, standard reference material analysis, and duplicate sample analysis by the analytical facility. Grain size analysis determinations were made according to the methods described by Folk (1980), which generally involves separation of the sand from the sediment sample, followed by pipette analysis of silt and clay.

Chemical residue analysis for the trace elements was performed by Environmental Trace Substances Research Center, Columbia, Missouri. Sediment and biota samples were analyzed for 20 trace elements using Inductively Coupled Plasma (ICP) emission spectroscopy, with preconcentration for selected elements for the biological samples. ICP measurements were made using a Jarrell-Ash Model 1100 Mark III spectrometer. Arsenic and selenium concentration determinations were made using hydride generation atomic absorption spectroscopy. Hydride generation atomic absorption measurements were made using a Perkin Elmer Model 603 spectrophotometer, with a Perkin-Elmer MHS-1 hydride generation accessory mounted for biological sample

analysis, and a Varian VGA-76 hydride generation accessory mounted for sediment sample analysis. Mercury concentration analysis was made using cold vapor atomic absorption. Cold vapor atomic absorption measurements were made using a Perkin-Elmer Model 403 spectrophotometer. The analytical detection limits for all the elements varied according to the particular analyte, and is presented with the tabulated data. The accuracy and precision of the chemical analysis was measured by method blank sample analysis, spike recovery sample analysis, standard reference material analysis, and duplicate sample analysis by the analytical facility. Quality assurance and quality control was monitored by the U.S. Fish and Wildlife Service, Patuxent Analytical Control Facility, Laurel, Maryland, and was reported as acceptable for all analytes.

Chemical residue analysis for the organic compounds was performed by Mississippi State Chemical Laboratory at Mississippi State University, Mississippi State, Mississippi. Sediment and biota samples were analyzed for aliphatic and aromatic hydrocarbons, organochlorine pesticides and total polychlorinated biphenyls (PCBs). Percent total volatile solids (TVS) determinations were performed on sediments by Environmental Trace Substances Research Center, Columbia, Missouri. Oil and grease determinations on sediments were made by Mississippi State Chemical Laboratory at Mississippi State University, Mississippi State, Mississippi.

Sediment samples were analyzed for 13 aliphatic hydrocarbons and 14 polycyclic aromatic hydrocarbons. The aliphatic and aromatic hydrocarbons were fractionated by initially extracting the sediment samples with acetone and petroleum ether. The hydrocarbon residues from the extraction were partitioned into petroleum ether, and then double-rinsed and concentrated prior to transfer to a silica gel column. Biota samples analyzed for aliphatic and aromatic hydrocarbons were initially digested in aqueous potassium hydroxide and extracted with methylene chloride, dried and reconstituted in petroleum ether prior to transfer to a silica gel column. The aliphatic fraction was separated from the hydrocarbon residue by eluting the aliphatics from the column with petroleum ether. The aromatic fraction was eluted from the silica gel column using methylene chloride/petroleum ether and methylene chloride, successively. The aliphatic fraction was then concentrated and quantified by capillary column flame ionization gas chromatography. The aromatic hydrocarbon fraction was concentrated, subjected to gel permeation chromatography for clean-up, and then

quantified by capillary column flame ionization gas chromatography and fluorescence high performance liquid chromatography.

Sediment samples were analyzed for 21 organochlorine pesticides and total PCBs. Sediment samples were initially extracted with acetone and hexane, and the residues partitioned into the hexane portion were washed, concentrated and then transferred to a Florisil mini-column, where the residues were eluted from the column into two primary elution fractions. Biota samples analyzed for organochlorine pesticides and PCBs were initially ground and mixed with anhydrous sodium sulfate, and then extracted in hexane using the soxhlet extraction method. The extract was concentrated to dryness for percent lipid determinations, and then extracted with acetone saturated with petroleum ether. Petroleum ether-partitioned concentrated residues were eluted in a Florisil column, and then separated into two fractions with diethyl ether/petroleum ether. Additional fractionation of PCBs from other organochlorines was performed by silicic acid column chromatography. Organochlorine and PCB residues in each of the fractions were quantified by packed column electron capture gas chromatography.

Percent total volatile solids (TVS) determinations were made for each sediment sample by weighing pre-weighed aliquots of oven-dried samples into fired crucibles, which were then fired at 550° C for a minimum of four hours. The crucible samples were cooled, desiccated and re-weighed, and the difference between the oven-dried aliquot and the fired sample, divided by the oven-dried sample weight, was used to determine the % total volatile solids for each sample. Oil and grease determinations were made by extracting a 50 gram sediment sample with acetone/petroleum ether three times, sequentially centrifuging, and then retaining the petroleum ether partitioned portion. This portion was washed with water, concentrated by Kuderna-Danish, and then transferred via petroleum ether rinsing through a bed of sodium sulphate to a tared glass tube, from which the percent oil and grease determinations were made after solvent removal under nitrogen.

For the organic analysis of sediments, the accuracy and precision of the chemical analysis was measured by reagent and matrix blank sample analysis, spike recovery sample analysis, and duplicate sample analysis, with confirmation of selected samples by gas chromatography/ mass spectrometry by the analytical facility. For the organic

analysis of biota samples, the accuracy was measured by reagent and matrix blank sample analysis and spike recovery with confirmation of selected samples by gas chromatography/ mass spectrometry by the analytical facility. The lower level of detection for the polycyclic aromatic hydrocarbon analysis was 0.01 ppm for both the sediment and tissue samples. The lower level of detection for the organochlorine compounds, with exception of toxaphene and PCBs, was 0.01 ppm for both sediment and tissue samples. The lower limit of detection for toxaphene and PCBs was 0.05 ppm. Quality assurance and quality control for biota and sediment sample analyses was monitored by the U.S. Fish and Wildlife Service Patuxent Analytical Control Facility, Laurel, Maryland, and was reported as acceptable for all analytes.

### Toxicity Tests

Static, chronic 10-day whole sediment toxicity tests using the amphipod H. azteca, were conducted with bed sediment samples collected from selected sites in the Yellowbank Slough drainage by personnel of the Minnesota Cooperative Fish and Wildlife Research Unit, Minneapolis, Minnesota (U.S. Fish and Wildlife Service 1991). Hyalella test organisms were obtained from laboratory reared cultures. Test conditions followed American Society for Testing and Materials (ASTM 1988) guidelines. Test organisms less than 3 mm were pipetted into 30 ml temporary holding beakers, which were then floated in 1000 ml experimental test beakers which contained 200 mls of sample bed sediment. The experimental test beakers were filled with well water to 1 liter total volume. After the water temperature in the temporary holding beakers containing the test organisms had equilibrated with the experimental test beaker water, the organisms were released below the water surface of the test chamber in the experimental test beakers. Organisms were fed laboratory rations of 250 um sieved rabbit pellets on days 0, 4 and 7 of the test. Dissolved oxygen, temperature, pH, hardness alkalinity and conductivity were measured at the start of the test prior to introduction of test organisms, and aeration and any observed mortality was recorded each day. At the end of the test on day 10, the test chamber contents were rinsed through a #40 U.S. Standard sieve. Remaining organisms were removed, counted, and any observed mortality recorded.

Static, acute 96-hour toxicity tests were also conducted with the cladoceran C. dubia

using surface water collected from the same sites bed sediments were sampled in the Yellowbank Slough region. The C. dubia bioassays were performed by the University of Iowa - Hygienics Laboratory in Des Moines, Iowa. Laboratory cultured organisms less than 24 hours old were placed in 100 ml beakers containing surface water samples collected the previous day from the field sampling locations. There were three replicates per sample, and each of the tests included a control (also with three replicates) using wellwater as the test solution for the control. Organisms were not fed for the duration of the test. A water sample was taken prior to introduction of the test organisms and analyzed for the following parameters: dissolved oxygen, temperature, pH, ammonia nitrogen, total ammonia, unionized ammonia, total hardness, total alkalinity, specific conductance and total residual chlorine. Dissolved oxygen, temperature and pH were monitored at 24-hour intervals after the start of the test, and again at the conclusion of the test for all replicates. Total mortality was calculated as number of organisms dead per number of organisms that were tested.

The Microtox™ Solid-Phase (Microbics Corp. 1991) and 100% Test Protocols (Microbics Corp. 1991) were used to conduct toxicity tests with bed sediment samples and pore water extractions both split with the aquatic invertebrate bulk sediment toxicity test samples, and samples collected from other sampling locations that were not assessed with the aquatic invertebrate toxicity tests. The Microtox tests have previously been shown to be sensitive indicators in the evaluation of oil drilling-associated contaminants (Stroscher 1980), as well as severely contaminated aquatic environments in general (Giesy et al. 1988). The Solid-Phase Test involves exposure of the luminescent marine bacterium P. phosphoreum directly to the solid phase of the material in a water suspension. Natural light production of this organism decreases when potentially toxic materials bound to sediment particles in the test sample interfere with the metabolism of the bacterium, and a reduction of light output by the organism is observed, theoretically in relation to the proportion of toxicity present in the sample. The 100% Assay Test Protocol is based on the same principle, with the exception that for the present study this procedure utilized pore water extracted from the bulk sediment samples as the test sample for exposure to the luminescent bacterium. Therefore, the major difference between the Solid-Phase and the 100% Assay Test Procedures is that the Solid-Phase Test is capable of detecting insoluble or soluble organic and inorganic toxic substances without initial sample extraction.

The Solid-Phase and 100% Assay Test procedures were conducted according to test protocols outlined by Microbics Corporation (1991). All tests were completed within two weeks of initial sample collections. The Solid-Phase and 100% Assay Tests were conducted using the Microtox Model 500 Analyzer, which quantifies reduction in light levels in the presence of increasing concentrations of a toxic substance in the sample, relative to a control which contains no sample material. The Solid-Phase Test conducted employed nine serial dilutions with an initial concentration of 10% (of the original sample material) and a dilution factor of 2. The 100% Assay Test Protocol employed 4 serial dilutions with an initial concentration of 91% and a dilution factor of 2. The assay time for each test was 20 minutes and the concentration units were reported in percentages with a 95% confidence range. No ionic adjustment was included for either test. After solid phase sample, pore water sample, and Microtox Reagent preparation, the tests were initiated and light levels were recorded automatically via computer with automatic data capture capability, using the Microtox computer software. Eight samples were analyzed for both the Solid-Phase and 100% Assay Test Protocols. Test data was automatically reduced, an EC value calculated, and a data report generated by the Microtox software program for each sample collected.



## Results

The results of the inorganic and organic analysis of bottom sediments collected from the Yellowbank Slough region were transformed to concentrations on a dry weight basis, based on the percent moisture determinations for each sample provided by the analytical facility, and are tabulated in Appendix A (Tables 3 through 7). Table 2(a) is a general summary of the analytical results for bed sediments for each site for all classes of chemical parameters analyzed: trace elements, aliphatic hydrocarbons, aromatic hydrocarbons, and organochlorine pesticides and PCBs. Table 3 shows the results of the total organic carbon and grain size analysis determinations for each site. One site in particular, YS-5, stands out as having a significantly higher percentage of sand in the sample that was collected. This site, as well as two other sites, YS-10 and YS-12, are all somewhat lower in the percent clay fraction, which theoretically is the fraction that may represent the highest proportion of adsorbed organics and inorganics, if present in substantial quantity. Sites YS-5 and YS-12 also have somewhat lower organic carbon percentages (on a dry weight basis), which should also affect contaminant adsorption. Sample YS-11 was actually collected at a crude oil spill site that was accidentally encountered during the 1990 field sampling, and has an elevated organic carbon content and no particle size distribution because the sample predominantly consisted of crude oil.

Table 4 shows the results of the trace element analysis of bed sediments collected from each of the sites. By comparison of concentrations of both negative controls (YS-1 and YS-12) and the crude oil positive control (YS-11), with the elements in samples collected from all other sites, six other elements appear to be significantly elevated in crude oil from this region: lead, copper, molybdenum, nickel, strontium, and zinc. Sites YS-2, YS-3, YS-7 and YS-9 consistently indicated very slight elevations of several of these inorganics (particularly copper and lead) with respect to the positive control and reference sites, which may be indicative of past or recent spills in these areas.

Table 5 shows the results of the aliphatic hydrocarbons scan. Aliphatic hydrocarbon analysis of the various sample sites shows a relationship between positive control and recently impacted sites, as well as which hydrocarbons are more prevalent in the

**Table 2(a). Summary of Yellowbank Slough 1990 bottom sediment chemical residue analysis results for trace elements, aliphatic hydrocarbons, polycyclic aromatic hydrocarbons, and organochlorines by general sampling location (data summarized from Appendix A). "Elevated" indicates the elevation of the majority of analytes of concern in crude oil in a particular parameter class over background sample residue analysis. "Not detected" indicates analytical parameters were not detected upon chemical analysis. "Background" indicates analytical parameters were significantly depressed with respect to positive control sample residue analysis.**

<b>Sample Location</b>	<b>Trace Elements</b>	<b>Aliphatic Hydrocarbons</b>	<b>Polycyclic Aromatic Hydrocarbons</b>	<b>Organo-chlorines</b>
<b>Upper Yellowbank Slough (YS-1)</b>	Not Elevated	Very Slightly Elevated	Not Elevated	Not Detected
<b>Middle Yellowbank Slough (YS-3)</b>	Not Elevated	Very Slightly Elevated	Not Elevated	Not Detected
<b>Lower Yellowbank Slough (YS-4)</b>	Not Elevated	Elevated	Slightly Elevated	Not Detected
<b>Goose Pond (YS-9)</b>	Not Elevated	Elevated	Slightly Elevated	Not Detected
<b>Middle Running Slough (YS-7)</b>	Very Slightly Elevated	Very Slightly Elevated	Not Elevated	Not Detected
<b>Lower Running Slough (YS-10)</b>	Not Elevated	Elevated	Elevated	Not Detected
<b>Shawneetown reference site (YS-12)</b>	Background	Background	Background	Background

crude oil found in that particular region of southeastern Illinois. Sample sites YS-4, YS-7, YS-9, and YS-10 were each sites at which recent spills were known or suspected to have occurred. Hydrocarbons which appear to be derived from crude oil spills are represented by each of the 13 aliphatics analyzed for with the exception of n-dodecane.

Table 6 shows the results of the polycyclic aromatic hydrocarbon analysis for bottom sediments, as well as percent total volatile solids, and oil and grease. The only aromatic hydrocarbon that appears to be present in significant concentrations in crude oil from the Yellowbank Slough region samples is phenanthrene. Polycyclic aromatic hydrocarbons are ubiquitous in the environment (Eisler 1987); many are typically found widely distributed at trace levels due to various anthropogenic sources associated with fossil fuel usage such as atmospheric deposition and stormwater runoff of combustion products, as well as due to biogenic sources. From the analytical data it can be concluded that the concentrations of phenanthrene detected at various locations on site are due principally to crude oil exposure though, because of its maximum elevation in the crude oil sample (YS-11) coupled with correspondingly lower elevations at a few of the other known spill sites (YS-4, YS-9, and YS-10).

Table 7 shows the results of the organochlorine pesticide and PCB analysis of bottom sediment samples. Organochlorine compounds and PCBs were not detected at any of the 1990 sampling sites in the Yellowbank Slough region, although trace concentrations of PCBs were detected at the off-site reference sampling location, YS-12.

With respect to the benthic invertebrates that were collected for chemical analysis, Tables 8 - 11 (Appendix B) show the results of the analysis for trace elements (Table 8), aliphatic hydrocarbons (Table 9), polycyclic aromatic hydrocarbons (Table 10), and organochlorine pesticides and PCBs (Table 11). The analytical results are presented both in mg/kg dry weight units [Tables 8(a), 9(a), 10(a), and 11(a), and in mg/kg wet weight units [Tables 8(b), 9(b), 10(b), and 11(b)]. The results of all chemical analysis of invertebrates are summarized in Table 2(b). Sites YS-9, Goose Pond, represents a biota sample (crayfish) from a known contaminated site. YS-1 represents a sample of mixed invertebrates from an upstream reference site. There were also two separate samples of mixed invertebrates (YS-12a) and crayfish (YS-12b). For the crayfish that were collected at site YS-9, several elements (aluminum,

**Table 2(b). Summary of Yellowbank Slough 1990 macroinvertebrate tissue chemical residue analysis results for trace elements, aliphatic hydrocarbons, polycyclic aromatic hydrocarbons, and organochlorines by general sampling location (data summarized from Appendix B). "Elevated" indicates the elevation of the majority of analytes of concern in crude oil in a particular parameter class over background sample residue analysis. "Not detected" indicates analytical parameters were not detected upon chemical analysis. "Background" indicates analytical parameters were significantly depressed with respect to contaminated sample residue analysis.**

<b>Sample Location</b>	<b>Trace Elements</b>	<b>Aliphatic Hydrocarbons</b>	<b>Polycyclic Aromatic Hydrocarbons</b>	<b>Organo-chlorines</b>
<b>Upper Yellowbank Slough (YS-1)</b>	Not Elevated	Very Slightly Elevated	Not Detected	Not Detected
<b>Goose Pond (YS-9)</b>	Elevated	Very Slightly Elevated	Not Elevated	Not Detected
<b>Shawneetown reference site (YS-12a)</b>	Background	Background	Not Detected	Not Detected
<b>Shawneetown reference site (YS-12b)</b>	Background	Background	Background	Not Detected

iron, barium, manganese, and zinc) in Table 8(a) appear to be elevated over the crayfish collected at YS-12. The mixed invertebrates in sample YS-12a all appear to be substantially elevated over the upstream reference sample collected at YS-1. Table 9(a) shows the results of the aliphatic hydrocarbon analysis of the invertebrate samples, and indicates that several hydrocarbons are elevated in crayfish at site YS-9 over background, but only slightly. Aliphatic hydrocarbons in other (mixed) benthic invertebrates do not appear to be elevated at all. Table 10(a) shows the results of the polycyclic aromatic hydrocarbon analysis of invertebrates; only two aromatics, phenanthrene and benzo(e)pyrene were detected in Goose Pond (YS-9) crayfish. Trace amounts were also detected in reference site (YS-12) crayfish. No aromatics were detected in the mixed benthic invertebrate samples collected from either reference site. Table 11(a) shows the results of the organochlorine pesticide and PCB analysis of invertebrates. No organochlorine compounds were detected in biota samples at any of the sites.

The results of the invertebrate species identification for each of the artificial substrates and benthos samples collected at Yellowbank Slough is shown in Table 12 (Appendix C). Sample YS-1 is the upstream reference site and YS-7 is a site where a suspected oil discharge had previously occurred. Generally, an increase in both species richness and diversity is reflected in both artificial substrates and the benthos sample at the reference site (YS-1), relative to the impacted site samples (YS-7).

During the 1991 field sampling season, surface water and bottom sediment samples were collected for laboratory toxicity tests, which were conducted with C. dubia and H. azteca. Bioassays were also conducted with P. phosphoreum using the Microtox™ toxicity testing system. The results of all laboratory toxicity tests are summarized in Table 2(c). Water quality samples were collected and analyzed in the field for five parameters for all sampling sites, and the results of the site water quality analysis is shown in Table 13 (Appendix D). Tables 14(a) and 14(b) (Appendix E) show the results of the 96-hour toxicity tests conducted with C. dubia, the results based on data from one control and four replicates. No toxicity was detected in surface water samples collected from four sites in the Yellowbank Slough region. Table 14(b) describes the water quality parameters monitored for the water samples tested at the start of the test. Table 15(a) (Appendix E) shows the results of the 10-day H. azteca toxicity tests conducted with bottom sediments collected from four sites in the

Table 2(c). Summary of the results of the toxicity testing conducted with Ceriodaphnia dubia, Hyalella azteca, and Microtox™ using surface water and bottom sediment samples collected from some general sampling locations in the Yellowbank Slough region in 1991 (data summarized from Appendices E and F).

Sample Location	<u>Ceriodaphnia dubia</u> 96-hour test	<u>Hyalella azteca</u> 10-day test	Microtox™ Tests:	
			Solid-Phase	100% Assay
Upper Yellowbank Slough (YS-1)	No toxicity	No toxicity	Moderately toxic	No data
Lower Yellowbank Slough (YS-4)	No toxicity	No toxicity	Moderately toxic	No toxicity
Middle Running Slough (YS-7)	No toxicity	No toxicity	Toxic	No toxicity
Goose Pond (YS-9)	No toxicity	No toxicity	Toxic	No toxicity
Shawneetown reference site (YS-12)	No toxicity	No toxicity	Slightly toxic	No toxicity

Yellowbank Slough area. Table 15(b) describes the water quality parameters that were measured in the test vessels at the start of the test. The toxicity observed was not significant for any of the four samples tested.

Table 16 (Appendix F) shows the results of the Microtox toxicity test conducted with bottom sediments and pore water collected from seven sites in the Yellowbank Slough region. While the pore water samples from the 100% Assay Test indicated no toxicity to the test bacterium, the Solid-Phase Test indicates significant toxicity for several of the samples collected. The lowest toxicity was observed with the off-site Shawneetown reference sample, YS-12. The Microtox data computer-generated reports for each set of tests are also provided in Appendix F.

## Discussion

According to the results of the bottom sediment sampling conducted in the Yellowbank Slough region, localized areas of contaminated sediments appear to be affected to the greatest extent by aliphatic hydrocarbons, and to lesser extent by polycyclic aromatic hydrocarbons, metals and other inorganics. Elevation of aliphatic hydrocarbons over background is very apparent at sampling sites YS-4, YS-6, YS-7, YS-9, YS-10 and YS-11, whereas aromatics are only slightly elevated at sites YS-4, YS-9, YS-10 and YS-11, and no metals are significantly elevated at any sampling location. Exposure to crude oil, which is comprised principally of aliphatic hydrocarbons, and to a much lesser extent, aromatic hydrocarbons, is the most likely cause for the concentrations observed at several of the sampling sites. The ecological risks of many of the aliphatics are comparatively less (in terms of toxicity) than those of polycyclic aromatics. Because many polycyclic aromatic hydrocarbons are carcinogenic and/or mutagenic, they have been studied and tested in greater detail.

Phenanthrene, which was the only aromatic hydrocarbon found in the crude oil sample and throughout the Yellowbank Slough region, is a three ring low molecular weight polycyclic aromatic hydrocarbon that is identified as a priority pollutant by the U.S. Environmental Protection Agency (Eisler 1987). Although it is in the low molecular weight class of PAHs that are generally rapidly metabolized when bioaccumulated, it does partition onto organics in sediments. It is characterized by an acute toxicity range of from 96 to 1150 ug/L (ppb) for freshwater aquatic invertebrates (U.S. Environmental Protection Agency 1991). The U.S. Environmental Protection Agency has proposed sediment quality criteria for phenanthrene for freshwater organisms based on the equilibrium partitioning approach with respect to sediment and pore water. Table 2(d) describes the sediment quality criteria for phenanthrene, which is presented on a micrograms of phenanthrene/ gram of organic carbon basis, as well as calculated specifically for two of the highest sediment phenanthrene concentrations detected in the Yellowbank Slough region during the present study, at sites YS-11 and YS-10. Compared with the phenanthrene concentrations on a dry weight basis (micrograms of phenanthrene/ gram of dry sediment) identified in bed sediments in the Yellowbank Slough region, the calculated sediment quality criteria are well above the maximum concentrations found



Table 2(d). Comparison of maximum concentrations (ug/g dry weight and ug/g<sub>oc</sub>) of phenanthrene detected in bed sediments in the Yellowbank Slough region, with local (on-site) background concentrations and available sediment quality criteria for the protection of benthic freshwater aquatic organisms.

Phenanthrene		
Maximum Concentrations Sampled:	Dry Weight	Organic Carbon Normalized
YS-11	9.86	57.32
YS-10	1.16	38.66
Background Concentrations:		
Local	Not detected	
Sediment Quality Criteria:	Dry Weight	Organic Carbon Normalized
YS-11	31.53 <sup>1</sup>	183.37 <sup>2</sup>
YS-10	5.50 <sup>1</sup>	
(General)	0.26 <sup>3</sup>	

<sup>1</sup>Calculated for the Yellowbank Slough region samples, based on 17.2% and 3.0% organic carbon for samples YS-11 and YS-10, respectively (U.S. EPA 1991).

<sup>2</sup>U.S. Environmental Protection Agency (1991); draft recommendation for the concentration of phenanthrene in sediments in freshwater aquatic systems that will not unacceptably affect benthic organisms (not taking into account potential contamination of higher trophic level organisms, or synergistic, additive or antagonistic effects of other chemicals present in the environment in addition to phenanthrene).

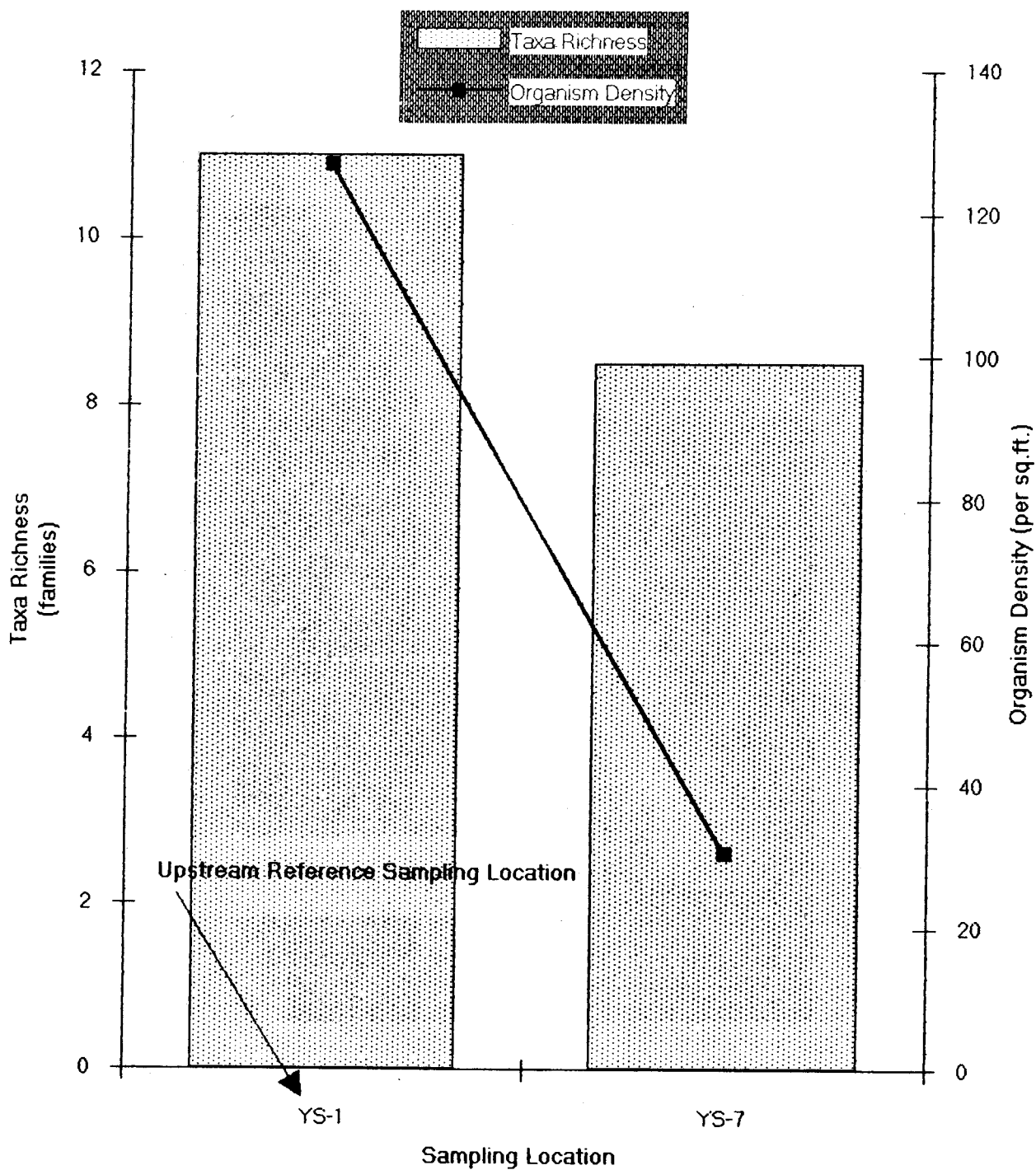
<sup>3</sup>Long and Morgan (1991); overall apparent effects threshold concentration of phenanthrene above which statistically significant biological effects occur for benthic organisms in general.

on site during the present study. The difference is much smaller for sediments having a lower organic carbon content (the site YS-10 phenanthrene concentration is 1.16 ug/g dry weight compared with the recommended quality criteria of 5.50 ug/g dry weight), which indicates that, in general, areas characterized by sediments having lower organic carbon content might be more severely impacted by phenanthrene exposure. Phenanthrene is soluble in water, and its solubility at 25° C (the ambient temperature on an average summer day) of 1.18 mg/L (ppm) (U.S. Environmental Protection Agency 1991) is much greater than its acute toxicity. Therefore, upon a release of crude oil, there still is a significant potential for adverse impacts to aquatic resources depending on the quantity spilled, based on the phenanthrene concentration in crude oil. (It is important to note here that the less than 1000 gram sample collected and analyzed during the present study might significantly underestimate the amount of phenanthrene present during a typical spill event, assuming a release of greater than one liter. The severity of impact of phenanthrene released to the environment under such circumstances would depend primarily on the quantity of crude oil released and the organic carbon content of the local bottom sediments.) Additionally, Long and Morgan (1991) describe an overall apparent effects threshold of 260 ppb (0.260 ppm) for phenanthrene in sediments [Table 2(d)]. This overall apparent effects threshold represents the sediment concentration above which statistically significant biological effects always occur, in terms of depressed benthos abundance and toxicity testing mortality, and therefore such effects may be anticipated in a given field situation.

The results of the chemical analysis of invertebrates generally did not correspond well with the bottom sediment chemical analysis. Of the inorganics that were found to be elevated in the crude oil sediment sample as well as at potentially impacted sites, only zinc was elevated in biota at these particular sites. The lack of accumulation of aliphatic and aromatic hydrocarbons in invertebrate tissue may be due to lack of capacity for some aquatic invertebrates to accumulate these hydrocarbons, when compared with some vertebrates (Eisler 1987; Hall and Coon 1988). However, phenanthrene and benzo(e)pyrene, which were also elevated in crude oil and sediments on site, were found to be elevated in crayfish samples.

As with other similar studies, it was anticipated that the effects of crude oil on biota might most effectively be evaluated with a combination of sediment chemistry and

biologically oriented approaches, such as species richness and diversity comparisons and invertebrate bioassays (Chapman et al. 1991). These two approaches were employed in the present study, in addition to the sediment chemistry. Analysis of species richness and diversity with artificial substrates reportedly provides a more reliable description of water (and habitat) quality conditions over a relatively long period of time (Modde and Drewes 1990), when compared with benthos samples which are only representative of macroinvertebrate assemblages at one point in time (U.S. Environmental Protection Agency 1990). Benthic invertebrate community health can be assessed generally by analyzing the species or taxa (genera/ families) richness in an aquatic community. The taxa richness, or variety of species, is anticipated to increase with corresponding increases in water and habitat quality, and habitat diversity (U.S. Environmental Protection Agency 1990). In the present study similar habitats were sampled for aquatic invertebrates (at sites YS-1 and YS-7), so this type of comparison can be made with minimal bias. Furthermore, even though artificial substrates tend to select for certain types of organisms (aquatic insects), comparison of artificial substrate collected data minimizes bias in bottom sampling due to physical habitat variation in water depth, light penetration, temperature differences and micro-habitat differences in bottom substrate types. Because taxa richness (at the family level) and numbers of individuals per species in the artificial substrate samples was found to be generally higher at the upstream reference site in the present study (shown in Figure 5), it may be concluded that in general, exposure to crude oil contaminants might adversely impact aquatic invertebrate community composition in the Yellowbank Slough region. Organism density per square foot calculated from the macroinvertebrate artificial substrate quantitative data obtained from the present study, yields an average density of 127 organisms per square foot for site YS-1 (the upstream reference), but only 30.5 organisms per square foot for site YS-7 (a recently impacted site). Over the long term, crude oil exposure might result in a reduced number of taxa and biomass of invertebrates in a given area, or at least a periodic reduction following and corresponding to periodic spill events. This same trend has been demonstrated with other similar studies (Albers et al. 1985; Shales et al. 1989; Woodward and Riley 1983), with as much as a 50% or greater reduction in benthic macroinvertebrates observed over a period of just a few weeks after a spill event. However, because a limited number of artificial substrates were placed and a limited number of benthic samples collected during the present study, this is an aspect that may need to be investigated further in the Yellowbank Slough study area.



**Figure 5. Bar graph of taxa richness comparisons for the artificial substrate aquatic macroinvertebrate colonization assemblages for sites YS-1 and YS-7 in the Yellowbank Slough region.**

Although the results of the Ceriodaphnia and Hyaella bioassays conducted were negative for the most part, the bioassays may also need to be investigated further. The bioassay results may be inconclusive, due to the manner in which the tests were conducted. Because metals were probably either not bioavailable or elevated enough to elicit a toxic response in the invertebrates tested, any adverse effects that might occur from crude oil exposure might be due exclusively to aliphatic and/or aromatic hydrocarbons. Some polycyclic aromatic hydrocarbons are known to be phototoxic in nature, and their toxic qualities are stimulated and enhanced by ultraviolet radiation from natural sunlight. In fact, photodegradation is considered to be one of the major chemical processes that control the short-term fate of oil, including crude oil, when releases occur in an aquatic environment (Robotham and Gill 1989). Those PAHs that have been tested in laboratory bioassays with invertebrates and vertebrates under conditions simulating natural sunlight include several commercially prepared pure compounds, as well as sediments characterized generally by PAH contamination (Davenport and Spacie 1991; Holst and Giesy 1989; Landrum et al. 1987; Oris and Giesy 1986; and Oris and Giesy 1987). The bioassays that were conducted during the present study were not conducted in a manner to evaluate any potential phototoxicity of polycyclic aromatic hydrocarbon crude oil components. Phenanthrene was the only aromatic hydrocarbon found in bottom sediments collected from the Yellowbank Slough region in significant amounts, and although this particular aromatic is not anticipated to be phototoxic in nature (Newsted and Giesy 1987), it has not been completely tested at the present time. Therefore, a re-analysis might provide different results. It should also be noted that the Microtox Solid-Phase test did detect small, but quantifiable differences in the toxicity of sediments collected on site, compared with the off-site reference location sample, with the more toxic Microtox samples representing areas known to have been impacted by recent spills.

The Yellowbank Slough region represents a diverse and generally productive habitat for wildlife species in general. However, current regional land use practices, specifically oil drilling-related activities, may not be compatible with wildlife management practices and objectives. Although drilling activities are state regulated, the continuous nature of oil exploration and extraction is what would primarily present a persistent threat of adverse impacts due to an oil spill. Significant oil spills were encountered in lotic portions of the study area by chance by the author on two separate occasions (one year apart) during relatively short-term site visits for field

sampling. Observations on these occasions strongly indicate that oil spills in the Yellowbank Slough region are a recurring event. Even the seasonal flooding that naturally occurs in the region might serve to redistribute sediments from localized areas impacted by crude oil spills, creating wider areas of crude oil impacted sediments. Table 2(e) is a summary of the trace elements, aliphatic hydrocarbons and polycyclic aromatic hydrocarbons that were generally found (based on the specific analytes which were originally analyzed for) in the Yellowbank Slough region, as derived from the crude oil sample YS-11. It also identifies those chemical analytes found at elevated concentrations in bed sediments at selected locations in the study area, which was indicative of past or recent localized releases of crude oil at those locations. Each ensuing future crude oil spill event in the Yellowbank Slough region would result in a release of (at least) these chemicals to the environment, in large or small quantities, depending upon the size of a particular release.

Although the aquatic invertebrate laboratory bioassay results of the present study indicate no adverse impacts potentially resulting from the release of crude oil, according to the results of other studies concerning the phototoxic nature of some aromatic hydrocarbons, the potential exists for significant impacts to aquatic resources to occur under different (more natural) circumstances (Landrum et al. 1987). Therefore, in spite of some of the conclusions of the results of the present study, crude oil spills could consistently result in significant impacts to aquatic invertebrate (Crunkilton and Duchrow 1990) and vertebrate (Hoffman and Albers 1984; Szaro 1977; Woodward et al. 1987) communities, and possibly thus indirectly affect the wildlife species that depend on aquatic invertebrates as a food source, or at least the quality of habitat available for those wildlife species.

If development of a region of wildlife habitat in cooperation with on-going industrial activities (such as oil extraction) is under consideration, then potential long-term impacts of recurring crude oil spills on wildlife habitat should first be very carefully considered. It is foreseeable that a contingency plan which would represent a mechanism for responding to oil spills will need to be developed in greater detail for a wildlife refuge with this type of industrial influence, than similar such current plans for other refuges in the national wildlife refuge system. Such a plan would need to identify physical and personnel resources locally available to help mitigate adverse impacts and minimize inevitable damage to natural resources. With the expectation

**Table 2(e). Summary of trace elements, aliphatic hydrocarbons, and polycyclic aromatic hydrocarbons generally found in crude oil from the Yellowbank Slough region at elevated concentrations (based on sample YS-11), and identified at substantially elevated (greater than one order of magnitude) concentrations at selected study area sampling sites in bed sediments during the present study.**

<b>Yellowbank Slough Region Crude Oil Chemical Composition</b>	<b>Substantially Elevated in Study Area</b>
<b>Trace Elements</b>	
copper lead* molybdenum nickel strontium zinc*	
<b>Aliphatic Hydrocarbons</b>	
n-dodecane*	X
n-tridecane*	X
n-tetradecane*	X
octylcyclohexane*	X
n-pentadecane*	X
nonylcyclohexane*	X
n-hexadecane*	X
n-heptadecane*	X
pristane*	X
n-octadecane*	X
phytane*	X
n-nonadecane*	X
n-eicosane*	X
<b>Polycyclic Aromatic Hydrocarbons</b>	
phenanthrene*	X

\*Elevation in crude oil greater than one order of magnitude over local background bed sediment concentrations.

that oil spills might occur with increased frequency in such an area, an inventory of specialized spill response equipment and supplies might need to be maintained on site by refuge personnel. It is also possible that certain refuge personnel would need to be permanently assigned special responsibilities with respect to actual spill response activities (such as preliminary environmental sampling and analysis, containment, countermeasures, clean-up, and disposal), as well as equipment/supplies maintenance. These responsibilities will also need to include wildlife dispersal, collection, cleaning and recovery expertise. Such duties for refuge personnel might require specialized personnel training at regular intervals. Also, a wildlife refuge with this type of industrial influence would be a prime candidate for more frequent than normal baseline monitoring for environmental contaminants.

In conclusion, a potentially significant increased cost of operation and maintenance of a wildlife refuge associated with this type of on-going industrial operation adjacent to (or even within) its boundaries is a real possibility. In the long term, this increased cost might well outweigh any benefits to wildlife resources derived from acquisition of such an area for wildlife habitat preservation.



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## Appendix A

### Analytical Chemistry Data: Bottom Sediments

Table 3. Total organic carbon and grain size analysis determinations for bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	% Moisture	TOC (% dry wt.)	% Sand	% Silt	% Clay
Upper Yellowbank Slough (upstream reference)	YS-1	1291.0	72.8	4.0	3.6	35.9	60.6
Upper Yellowbank Slough	YS-2	1366.0	73.5	4.6	3.3	35.2	61.5
Middle Yellowbank Slough	YS-3	1332.0	71.6	4.2	4.0	35.5	60.4
Lower Yellowbank Slough	YS-4	839.9	45.0	3.0	1.9	53.3	44.8
Lower Yellowbank Slough	YS-5	1437.0	47.4	1.8	29.9	39.0	31.0
Lower Running Slough	YS-6	813.1	40.7	2.7	2.3	54.8	42.9
Middle Running Slough	YS-7	1430.0	58.7	3.1	1.9	40.7	57.3
Middle Running Slough	YS-8	914.5	58.5	2.5	14.6	41.4	44.0
Goose Pond	YS-9	636.5	49.7	3.0	4.2	52.2	43.6
Lower Running Slough	YS-10	1409.0	42.4	3.0	1.7	67.5	30.7
Crude Oil Spill Site (positive control)	YS-11	1410.0	22.7	17.2	<sup>1</sup> ***	***	***
Shawneetown Vicinity (negative control)	YS-12	1325.0	32.2	1.8	23.7	46.2	30.1

<sup>1</sup>Analysis not conducted due to very high organic content.

Table 4. Concentrations (mg/kg dry weight) of trace elements detected in sediments collected from the Yellowbank Slough region in 1990. (Nominal analytical detection limits are provided at the end of the table.)

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Al	As	Ba	Be	B	Cd	Cr
Upper Yellowbank Slough (upstream reference)	YS-1	1323.0	73.9	38700.0	6.8	226.0	1.6	3.0	0.4	36.0
Upper Yellowbank Slough	YS-2	792.4	69.1	37600.0	10.2	260.0	1.7	3.0	0.6	35.0
Middle Yellowbank Slough	YS-3	1289.0	73.0	46600.0	8.1	288.0	1.8	5.0	0.7	43.0
Lower Yellowbank Slough	YS-4	1024.0	47.7	37900.0	9.5	224.0	1.6	5.0	0.6	39.0
Lower Yellowbank Slough	YS-5	1458.0	52.5	27700.0	10.6	179.0	1.3	3.0	0.4	29.0
Lower Running Slough	YS-6	1000.0	49.4	30100.0	7.4	204.0	1.4	4.0	0.6	31.0
Middle Running Slough	YS-7	832.3	57.2	37000.0	12.6	262.0	1.7	3.0	0.7	36.0
Middle Running Slough	YS-8	1474.0	64.5	23400.0	8.0	209.0	1.5	3.0	0.4	25.0
Goose Pond	YS-9	1314.0	64.7	35800.0	6.1	246.0	1.6	5.0	0.5	37.0
Lower Running Slough	YS-10	1411.0	47.0	24700.0	7.7	164.0	1.2	4.0	0.6	27.0
Crude Oil Spill Site (positive control)	YS-11	1612.0	21.9	25400.0	7.2	197.0	1.2	4.0	0.5	37.0
Shawneetown Vicinity (negative control)	YS-12	1285.0	33.8	23000.0	12.7	190.0	1.5	4.0	0.4	27.0
Analytical Detection Limits				3.0	0.1	0.1	0.1	2.0	0.3	1.0

Table 4 (cont.). Concentrations (mg/kg dry weight) of trace elements detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Cu	Fe	Pb	Mg	Mn	Hg	Mo
Upper Yellowbank Slough (upstream reference)	YS-1	25.4	40000.0	26.0	4520.0	1120.0	0.075	<1.0
Upper Yellowbank Slough	YS-2	28.3	41700.0	33.0	4690.0	904.0	0.069	<1.0
Middle Yellowbank Slough	YS-3	28.4	41300.0	30.0	5440.0	1010.0	0.075	<1.0
Lower Yellowbank Slough	YS-4	24.6	36300.0	26.0	5410.0	669.0	0.083	<1.0
Lower Yellowbank Slough	YS-5	19.8	32500.0	20.0	5450.0	570.0	0.063	<1.0
Lower Running Slough	YS-6	23.7	34300.0	25.0	4990.0	804.0	0.074	<1.0
Middle Running Slough	YS-7	27.2	46400.0	31.0	4910.0	1630.0	0.083	<1.0
Middle Running Slough	YS-8	22.7	34200.0	26.0	3600.0	1260.0	0.069	<1.0
Goose Pond	YS-9	22.3	33000.0	28.0	4730.0	667.0	0.078	<1.0
Lower Running Slough	YS-10	20.8	27700.0	22.0	5220.0	622.0	0.069	<1.0
Crude Oil Spill Site (positive control)	YS-11	55.7	38600.0	110.0	3470.0	604.0	0.069	3.0
Shawneetown Vicinity (negative control)	YS-12	18.0	36000.0	29.0	3760.0	1170.0	0.063	<1.0
Analytical Detection Limits		0.2	10.0	6.0	4.0	0.2	0.01	1.0



Table 4 (cont.). Concentrations (mg/kg dry weight) of trace elements detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Ni	Se	Ag	Sr	Tl	V	Zn
Upper Yellowbank Slough (upstream reference)	YS-1	30.0	0.76	2.0	33.1	<5.0	40.0	120.0
Upper Yellowbank Slough	YS-2	33.0	0.71	2.0	28.1	<5.0	42.0	123.0
Middle Yellowbank Slough	YS-3	36.0	0.66	2.0	37.6	<5.0	54.0	130.0
Lower Yellowbank Slough	YS-4	33.0	0.50	<2.0	30.7	<5.0	52.2	115.0
Lower Yellowbank Slough	YS-5	28.0	0.40	<2.0	30.3	<5.0	36.0	90.2
Lower Running Slough	YS-6	31.0	0.40	<2.0	28.0	<5.0	39.0	109.0
Middle Running Slough	YS-7	35.0	0.50	2.0	33.9	<5.0	43.0	128.0
Middle Running Slough	YS-8	28.0	0.40	<2.0	23.2	<5.0	27.0	98.6
Goose Pond	YS-9	30.0	0.40	<2.0	38.2	<5.0	46.0	103.0
Lower Running Slough	YS-10	25.0	0.50	<2.0	24.0	<5.0	36.0	93.1
Crude Oil Spill Site (positive control)	YS-11	50.0	0.20	2.0	81.7	<5.0	37.0	259.0
Shawneetown Vicinity (negative control)	YS-12	32.0	0.40	<2.0	24.2	<5.0	37.0	97.5
Analytical Detection Limits		1.0	0.2	2.0	0.1	5.0	0.5	0.5

Table 5. Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	% Moisture	n-Dodecane	n-Tridecane	n-Tetradecane	Octylcyclohexane
Upper Yellowbank Slough (upstream reference)	YS-1	853.0	75.8	ND <sup>1</sup>	ND	0.04	ND
Upper Yellowbank Slough	YS-2	773.0	77.6	ND	ND	0.04	ND
Middle Yellowbank Slough	YS-3	798.0	74.6	0.03	0.03	0.03	ND
Lower Yellowbank Slough	YS-4	1010.0	496.0	0.31	0.25	0.65	0.47
Lower Yellowbank Slough	YS-5	825.0	68.6	0.03	0.03	0.06	ND
Lower Running Slough	YS-6	1010.0	46.8	0.03	0.07	0.11	0.05
Middle Running Slough	YS-7	748.0	59.0	0.02	0.04	0.09	0.02
Middle Running Slough	YS-8	756.0	69.6	0.03	0.03	0.03	ND
Goose Pond	YS-9	865.0	73.6	3.37	5.6	6.8	1.89
Lower Running Slough	YS-10	1080.0	52.0	3.5	13.5	22.9	10.6
Crude Oil Spill Site (positive control)	YS-11	687.0	24.0	ND	43.4	68.4	92.1
Shawneetown Vicinity (negative control)	YS-12	568.0	30.6	ND	ND	0.01	ND

<sup>1</sup>ND = None detected.

Table 5 (cont.). Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Pentadecane	Monylcyclohexane	n-Hexadecane	n-Heptadecane	Pristane
Upper Yellowbank Slough (upstream reference)	YS-1	0.08	ND	0.04	0.61	0.41
Upper Yellowbank Slough	YS-2	0.04	ND	0.08	0.31	0.26
Middle Yellowbank Slough	YS-3	0.07	ND	0.07	0.19	0.15
Lower Yellowbank Slough	YS-4	0.83	1.01	1.28	2.7	4.1
Lower Yellowbank Slough	YS-5	0.09	0.06	0.09	0.19	0.31
Lower Running Slough	YS-6	0.15	0.07	0.16	0.22	0.30
Middle Running Slough	YS-7	0.14	0.04	0.14	0.31	0.24
Middle Running Slough	YS-8	0.03	ND	0.03	0.16	0.09
Goose Pond	YS-9	9.09	2.80	9.09	9.09	7.95
Lower Running Slough	YS-10	37.5	18.1	37.5	35.4	56.2
Crude Oil Spill Site (positive control)	YS-11	101.3	157.8	101.3	90.7	723.6
Shawneetown Vicinity (negative control)	YS-12	0.01	ND	0.02	0.12	0.05

Table 5 (cont.). Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Octadecane	Phytane	n-Nonadecane	n-Eicosane
Upper Yellowbank Slough (upstream reference)	YS-1	0.16	0.90	0.2	0.08
Upper Yellowbank Slough	YS-2	0.13	0.40	0.22	0.08
Middle Yellowbank Slough	YS-3	0.07	0.23	0.39	0.07
Lower Yellowbank Slough	YS-4	1.4	2.9	1.21	1.6
Lower Yellowbank Slough	YS-5	0.12	0.25	0.12	0.12
Lower Running Slough	YS-6	0.16	0.18	0.15	0.15
Middle Running Slough	YS-7	0.14	0.19	0.17	0.17
Middle Running Slough	YS-8	0.09	0.16	0.19	0.09
Goose Pond	YS-9	6.81	4.54	6.4	5.6
Lower Running Slough	YS-10	31.25	31.25	22.9	27.08
Crude Oil Spill Site (positive control)	YS-11	97.3	434.2	65.7	90.7
Shawneetown Vicinity (negative control)	YS-12	0.02	0.02	0.02	0.05

Table 6. Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs), percent total volatile solids, and oil and grease (ppm) detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Napthalene	Fluorene	Phenanthrene	Anthracene
Upper Yellowbank Slough (upstream reference)	YS-1	853.0	75.8	ND <sup>1</sup>	ND	ND	ND
Upper Yellowbank Slough	YS-2	773.0	77.6	ND	ND	ND	ND
Middle Yellowbank Slough	YS-3	798.0	74.6	ND	ND	ND	ND
Lower Yellowbank Slough	YS-4	1010.0	49.6	ND	0.03	0.27	ND
Lower Yellowbank Slough	YS-5	825.0	68.6	ND	ND	ND	ND
Lower Running Slough	YS-6	1010.0	46.8	ND	ND	0.03	ND
Middle Running Slough	YS-7	748.0	59.0	ND	ND	ND	ND
Middle Running Slough	YS-8	756.0	69.6	ND	ND	ND	ND
Goose Pond	YS-9	865.0	73.6	ND	0.03	0.30	ND
Lower Running Slough	YS-10	1080.0	52.6	ND	ND	1.16	0.104
Crude Oil Spill Site (positive control)	YS-11	687.0	24.0	0.07	ND	9.86	ND
Shawneetown Vicinity (negative control)	YS-12	568.0	30.6	ND	ND	0.01	ND

<sup>1</sup>ND = None detected.

Table 6 (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs), percent total volatile solids, and oil and grease (ppm) detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Fluoranthrene	Pyrene	1,2-benzanthracene	Chrysene	Benzo(b)flouranthrene
Upper Yellowbank Slough (upstream reference)	YS-1	0.04	ND	ND	ND	0.04
Upper Yellowbank Slough	YS-2	ND	ND	ND	ND	0.04
Middle Yellowbank Slough	YS-3	ND	ND	ND	0.07	0.03
Lower Yellowbank Slough	YS-4	0.15	0.07	0.03	ND	0.57
Lower Yellowbank Slough	YS-5	ND	ND	ND	ND	0.03
Lower Running Slough	YS-6	0.03	0.02	0.02	0.02	0.02
Middle Running Slough	YS-7	ND	ND	ND	ND	0.03
Middle Running Slough	YS-8	ND	ND	ND	ND	0.06
Goose Pond	YS-9	ND	0.03	0.07	0.07	0.03
Lower Running Slough	YS-10	ND	ND	0.64	ND	ND
Crude Oil Spill Site (positive control)	YS-11	ND	ND	ND	ND	ND
Shawneetown Vicinity (negative control)	YS-12	0.01	ND	0.01	0.02	0.01

Table 6 (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs), percent total volatile solids, and oil and grease (ppm) detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Benzo(k)fluoranthrene	Benzo(e)pyrene	Benzo(a)pyrene	1,2,5,6-dibenzanthracene
Upper Yellowbank Slough (upstream reference)	YS-1	ND	ND	0.04	ND
Upper Yellowbank Slough	YS-2	ND	ND	ND	ND
Middle Yellowbank Slough	YS-3	ND	0.15	ND	ND
Lower Yellowbank Slough	YS-4	0.01	ND	0.05	ND
Lower Yellowbank Slough	YS-5	ND	ND	ND	ND
Lower Running Slough	YS-6	ND	0.03	0.02	ND
Middle Running Slough	YS-7	ND	0.04	ND	ND
Middle Running Slough	YS-8	ND	ND	ND	ND
Goose Pond	YS-9	ND	0.03	ND	ND
Lower Running Slough	YS-10	ND	ND	ND	ND
Crude Oil Spill Site (positive control)	YS-11	ND	ND	ND	ND
Shawneetown Vicinity (negative control)	YS-12	ND	0.01	0.01	ND

Table 6 (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs), percent total volatile solids, and oil and grease (ppm) detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Benzo(g,h,i)perylene	Percent Total Volatile Solids	Oil and Grease (ppm)
Upper Yellowbank Slough (upstream reference)	YS-1	ND	15.3	410
Upper Yellowbank Slough	YS-2	ND	16.3	558
Middle Yellowbank Slough	YS-3	ND	16.2	604
Lower Yellowbank Slough	YS-4	0.41	11.3	5100
Lower Yellowbank Slough	YS-5	ND	8.3	552
Lower Running Slough	YS-6	0.02	9.7	500
Middle Running Slough	YS-7	ND	12.0	508
Middle Running Slough	YS-8	0.06	10.8	350
Goose Pond	YS-9	ND	11.4	962
Lower Running Slough	YS-10	ND	9.5	8700
Crude Oil Spill Site (positive control)	YS-11	ND	34.7	25300
Shawneetown Vicinity (negative control)	YS-12	ND	7.2	120



Table 7. Concentrations (mg/kg dry weight) of organochlorine compounds detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	HCB	Alpha BHC	Beta BHC	Gamma BHC	Delta BHC	Heptachlor Epoxide
Upper Yellowbank Slough (upstream reference)	YS-1	853.0	75.8	ND <sup>1</sup>	ND	ND	ND	ND	ND
Upper Yellowbank Slough	YS-2	773.0	77.6	ND	ND	ND	ND	ND	ND
Middle Yellowbank Slough	YS-3	798.0	74.6	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-4	1010.0	49.6	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-5	825.0	68.6	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-6	1010.0	46.8	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-7	748.0	59.0	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-8	756.0	69.6	ND	ND	ND	ND	ND	ND
Goose Pond	YS-9	865.0	73.6	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-10	1080.0	52.0	ND	ND	ND	ND	ND	ND
Crude Oil Spill Site (positive control)	YS-11	687.0	24.0	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (negative control)	YS-12	568.0	30.6	ND	ND	ND	ND	ND	ND

<sup>1</sup>ND = None detected.

Table 7 (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Oxy-chlordane	Alpha-chlordane	Gamma-chlordane	Cis-Monachlor	Trans-Monachlor	Dieldrin	Endrin	Mirex
Upper Yellowbank Slough (upstream reference)	YS-1	ND	ND	ND	ND	ND	0.04	ND	ND
Upper Yellowbank Slough	YS-2	ND	ND	ND	ND	ND	ND	ND	ND
Middle Yellowbank Slough	YS-3	ND	ND	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-4	ND	ND	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-5	ND	ND	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-6	ND	ND	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-7	ND	ND	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-8	ND	ND	ND	ND	ND	ND	ND	ND
Goose Pond	YS-9	ND	ND	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-10	ND	ND	ND	ND	ND	ND	ND	ND
Crude Oil Spill Site (positive control)	YS-11	ND	ND	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (negative control)	YS-12	ND	ND	ND	ND	ND	ND	ND	ND

Table 7 (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	o,p'DDE	p,p'DDE	o,p'DDD	p,p'DDD	o,p'DDT	p,p'DDT
Upper Yellowbank Slough (upstream reference)	YS-1	ND	ND	ND	ND	ND	ND
Upper Yellowbank Slough	YS-2	ND	ND	ND	ND	ND	ND
Middle Yellowbank Slough	YS-3	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-4	ND	ND	ND	ND	ND	ND
Lower Yellowbank Slough	YS-5	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-6	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-7	ND	ND	ND	ND	ND	ND
Middle Running Slough	YS-8	ND	ND	ND	ND	ND	ND
Goose Pond	YS-9	ND	ND	ND	ND	ND	ND
Lower Running Slough	YS-10	ND	ND	ND	ND	ND	ND
Crude Oil Spill Site (positive control)	YS-11	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (negative control)	YS-12	ND	ND	ND	ND	ND	ND

Table 7 (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in bottom sediments collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Total PCB's	Toxaphene
Upper Yellowbank Slough (upstream reference)	YS-1	0.87	ND
Upper Yellowbank Slough	YS-2	ND	ND
Middle Yellowbank Slough	YS-3	ND	ND
Lower Yellowbank Slough	YS-4	ND	ND
Lower Yellowbank Slough	YS-5	ND	ND
Lower Running Slough	YS-6	ND	ND
Middle Running Slough	YS-7	ND	ND
Middle Running Slough	YS-8	ND	ND
Goose Pond	YS-9	ND	ND
Lower Running Slough	YS-10	ND	ND
Crude Oil Spill Site (positive control)	YS-11	ND	ND
Shawneetown Vicinity (negative control)	YS-12	0.17	ND

## **Appendix B**

### **Analytical Chemistry Data: Aquatic Macroinvertebrates**

Table 8(a). Concentrations (mg/kg dry weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.  
(Nominal analytical detection limits are provided at the end of the table.)

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Al	As	Ba	Be	B	Cd	Cr
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	61.7	1780.0	0.93	48.0	0.057	<10.0	0.06	1.7
Goose Pond Crayfish	YS-9	221.0	76.7	1570.0	4.4	235.0	0.083	<10.0	0.27	2.0
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	48.4	4290.0	2.7	153.0	0.11	<10.0	0.09	3.8
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	77.3	53.7	0.40	32.0	<0.01	<9.0	0.04	0.3
Analytical Detection Limits				0.5	0.1	0.5	0.01	10.0	1.0	0.1

Table 8(a) (cont.). Concentrations (mg/kg dry weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Cu	Fe	Pb	Mg	Mn	Hg	Mo
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	9.17	2060.0	2.2	318.0	675.0	0.016	<5.0
Goose Pond Crayfish	YS-9	34.1	4970.0	5.9	2350.0	1180.0	0.29	<5.0
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	24.5	4210.0	2.5	1030.0	905.0	0.035	<5.0
Shawneetown Vicinity (reference site) Crayfish	YS-12b	45.7	110.0	2.0	2800.0	18.0	0.10	<5.0
Analytical Detection Limits		0.04	2.0	0.7	2.0	0.05	0.005	5.0

Table 8(a) (cont.). Concentrations (mg/kg dry weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Ni	Se	Ag	Sr	Tl	V	Zn
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	4.3	0.09	<10.0	133.0	<0.6	3.0	13.8
Goose Pond Crayfish	YS-9	3.5	0.44	<10.0	245.0	<0.6	3.0	396.0
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	12.0	0.35	<10.0	229.0	<0.8	7.0	29.7
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.87	0.36	<9.0	2110.0	<0.6	<2.0	65.8
Analytical Detection Limits		0.2	0.05	10.0	0.7	0.6	2.0	0.06



Table 8(b). Concentrations (mg/kg wet weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Al	As	Ba	Be	B	Cd	Cr
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	61.7	681.7	0.356	18.3	0.021	3.8	0.022	0.651
Goose Pond Crayfish	YS-9	221.0	76.7	365.8	1.02	54.7	0.019	2.33	0.062	0.466
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	48.4	2213.6	1.39	78.9	0.056	<5.16	0.046	1.96
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	77.3	12.1	0.09	7.26	0.002	2.04	0.009	0.06

Table 8(b) (cont.). Concentrations (mg/kg wet weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Cu	Fe	Pb	Mg	Mn	Hg	Mo
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	3.51	788.9	0.842	121.7	258.5	0.006	1.9
Goose Pond Crayfish	YS-9	7.94	1158.0	1.37	547.5	274.9	0.06	1.16
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	12.64	2172.0	1.29	531.4	466.9	0.018	2.58
Shawneetown Vicinity (reference site) Crayfish	YS-12b	10.3	24.97	0.454	635.6	4.08	0.02	1.13

Table 8(b) (cont.). Concentrations (mg/kg wet weight) of trace elements detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Ni	Se	Ag	Sr	Tl	V	Zn
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	1.64	0.03	3.83	50.93	<0.22	1.14	5.28
Goose Pond Crayfish	YS-9	0.815	0.10	2.33	57.0	0.139	0.69	92.2
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	6.19	0.18	<5.16	118.16	0.412	3.61	15.32
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.197	0.08	2.04	478.9	0.136	0.45	14.9

Table 9(a). Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Percent Lipid	n-Dodecane	n-Tridecane	n-Tetradecane	Octylcyclohexane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	0.16	ND <sup>1</sup>	ND	0.04	ND
Goose Pond Crayfish	YS-9	221.0	82.5	0.78	0.17	0.17	0.28	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	0.48	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	1.14	0.09	ND	0.14	ND

<sup>1</sup>ND = None detected.

Table 9(a) (cont.). Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Pentadecane	Nonylcyclohexane	n-Hexadecane	n-Heptadecane	Pristane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	0.04	ND	0.06	0.21	ND
Goose Pond Crayfish	YS-9	0.28	0.171	0.34	0.40	1.48
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	0.08	ND	0.05	0.08	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.14	ND	ND	1.90	ND

Table 9(a) (cont.). Concentrations (mg/kg dry weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Octadecane	Phytane	n-Nonadecane	n-Eicosane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	0.06	0.04	ND	0.09
Goose Pond Crayfish	YS-9	0.34	1.37	0.28	0.74
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	0.054	0.14
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	0.19

Table 9(b). Concentrations (mg/kg wet weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Percent Lipid	n-Dodecane	n-Tridecane	n-Tetradecane	Octylcyclohexane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	0.16	ND <sup>1</sup>	ND	0.02	ND
Goose Pond Crayfish	YS-9	221.0	82.5	0.78	0.03	0.03	0.05	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	0.48	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	1.14	0.02	ND	0.03	ND

<sup>1</sup>ND = None detected.

Table 9(b) (cont.). Concentrations (mg/kg wet weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Pentadecane	Nonylcyclohexane	n-Hexadecane	n-Heptadecane	Pristane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	0.02	ND	0.03	0.09	ND
Goose Pond Crayfish	YS-9	0.05	0.03	0.06	0.07	0.26
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	0.05	ND	0.03	0.05	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.03	ND	ND	0.4	ND



Table 9(b) (cont.). Concentrations (mg/kg wet weight) of aliphatic hydrocarbons detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	n-Octadecane	Phytane	n-Monadecane	n-Eicosane
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	0.03	0.02	ND	0.04
Goose Pond Crayfish	YS-9	0.06	0.24	0.05	0.13
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	0.03	0.08
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	0.04

Table 10(a). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Napthalene	Fluorene	Phenanthrene	Anthracene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	ND <sup>1</sup>	ND	ND	ND
Goose Pond Crayfish	YS-9	221.0	82.5	ND	ND	0.05	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	ND	ND	0.09	ND

<sup>1</sup>ND = None detected.

Table 10(a) (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Fluoranthrene	Pyrene	1,2-benzanthracene	Chrysene	Benzo(b)fluoranthrene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.09	0.04	ND	ND	0.04

Table 10(a) (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Benzo(k)fluoranthrene	Benzo(e)pyrene	Benzo(a)pyrene	1,2,5,6-dibenzanthracene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	0.05	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	0.04	ND

Table 10(a) (cont.). Concentrations (mg/kg dry weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Benzo(g,h,i)perylene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND
Goose Pond Crayfish	YS-9	ND
Shawneetown (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND
Shawneetown (reference site) Crayfish	YS-12b	ND

Table 10(b). Concentrations (mg/kg wet weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	Napthalene	Fluorene	Phenanthrene	Anthracene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	ND <sup>1</sup>	ND	ND	ND
Goose Pond Crayfish	YS-9	221.0	82.5	ND	ND	0.01	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	ND	ND	0.02	ND

<sup>1</sup>ND = None detected.

Table 10(b) (cont.). Concentrations (mg/kg wet weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Fluoranthrene	Pyrene	1,2-benzanthracene	Chrysene	Benzo(b)fluoranthrene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	0.02	0.01	ND	ND	0.01

Table 10(b) (cont.). Concentrations (mg/kg wet weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Benzo(k)fluoranthrene	Benzo(e)pyrene	Benzo(a)pyrene	1,2,5,6-dibenzanthracene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	0.01	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	0.01	ND



Table 10(b) (cont.). Concentrations (mg/kg wet weight) of polycyclic aromatic hydrocarbons (PAHs) detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

<u>Sample Location</u>	<u>Sample Number</u>	<u>Benzo(g,h,i)perylene</u>
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND
Goose Pond Crayfish	YS-9	ND
Shawneetown (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND
Shawneetown (reference site) Crayfish	YS-12b	ND

Table 11(a). Concentrations (mg/kg dry weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	HCB	Alpha BHC	Beta BHC	Gamma BHC	Delta BHC	Heptachlor Epoxide
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	ND <sup>1</sup>	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	221.0	82.5	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	ND	ND	ND	ND	ND	ND

<sup>1</sup>ND = None detected.

Table 11(a) (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Oxy-chlordane	Alpha-chlordane	Gamma-chlordane	Cis-Nonachlor	Trans-Nonachlor	Dieldrin	Endrin	Mirex
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Invertebrates	YS-12a	ND	ND	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	ND	ND	ND	ND	ND

Table 11(a) (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	o,p'DDE	p,p'DDE	o,p'DDD	p,p'DDD	o,p'DDT	p,p'DDT
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	ND	ND	ND

Table 11(a) (cont.). Concentrations (mg/kg dry weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Total PCB's	Toxaphene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND
Goose Pond Crayfish	YS-9	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND

Table 11(b). Concentrations (mg/kg wet weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Sample Weight(g)	Percent Moisture	HCB	Alpha BHC	Beta BHC	Gamma BHC	Delta BHC	Heptachlor Epoxide
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	402.0	57.0	ND <sup>1</sup>	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	221.0	82.5	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	195.0	44.0	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	139.0	79.0	ND	ND	ND	ND	ND	ND

<sup>1</sup>ND = None detected.

Table 11(b) (cont.). Concentrations (mg/kg wet weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Oxy-chlordane	Alpha-chlordane	Gamma-chlordane	Cis-Monachlor	Trans-Monachlor	Dieldrin	Endrin	Mirex
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Inveretbrates	YS-12a	ND	ND	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	ND	ND	ND	ND	ND

Table 11(b) (cont.). Concentrations (mg/kg wet weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	o,p'DDE	p,p'DDE	o,p'DDD	p,p'DDD	o,p'DDT	p,p'DDT
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND	ND	ND	ND	ND
Goose Pond Crayfish	YS-9	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND	ND	ND	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND	ND	ND	ND	ND



Table 11(b) (cont.). Concentrations (mg/kg wet weight) of organochlorine compounds detected in macroinvertebrates collected from the Yellowbank Slough region in 1990.

Sample Location	Sample Number	Total PCB's	Toxaphene
Upper Yellowbank Slough (upstream reference site) Benthic Aquatic Macroinvertebrates	YS-1	ND	ND
Goose Pond Crayfish	YS-9	ND	ND
Shawneetown Vicinity (reference site) Benthic Aquatic Macroinvertebrates	YS-12a	ND	ND
Shawneetown Vicinity (reference site) Crayfish	YS-12b	ND	ND

## **Appendix C**

### **Aquatic Macroinvertebrate Diversity Data**

Table 12. Natural and artificial substrate aquatic macroinvertebrate diversity data for Yellowbank Slough in 1990.

Sample Location: YS-1 (upstream reference)

	Benthic Sediment Samples	Artificial Substrate 1	Artificial Substrate 2
Annelida			
Oligochaeta			
Haplotaxida			
Tubificidae			16
Hirudinea			
Rhynchobdellida		7	8
Glossiphoniidae			
Pharyngobdellida			1
Erpobdellidae			
Mollusca			
Gastropoda			
Basommatophora		1	4
Hydrobidae		3	
Lymnaeidae			
Planorbidae		2	
Physidae			
Pelecypoda			
Heterodonta		1	6
Pisidiidae			
Arthropoda			
Crustacea			
Amphipoda		40	76
Talitridae	7		
Insecta			
Coleoptera			
Dytiscidae			
Haliplidae			
Helodidae		2	
Hydrophilidae			
Diptera			
Chironomidae			
Chironominae	42	10	14
Tanypodinae	13	1	3
Ceratopogonidae	10		1
Culicidae (Chaoborinae)	336		
Empidae			
Stratiomyidae	2		
Ephemeroptera			
Baetidae	4		
Caenidae	1	1	2
Heptageniidae	2		
Hemiptera			
Belostomatidae	1		
Corixidae	2		
Lepidoptera			
Pyrallidae	1		
Odonata (Anisoptera)			
Corduliidae	1		
Libellulidae	1	10	14
Odonata (Zygoptera)			
Coenagrionidae	3	9	22

Table 12 (cont.). Natural and artificial substrate aquatic macroinvertebrate diversity data for Yellowbank Slough in 1990.

Sample Location: YS-7

	Benthic Sediment Samples	Artificial Substrate 1	Artificial Substrate 2
Annelida			
Oligochaeta			
Haplotaxida			
Tubificidae	1		14
Hirudinea			
Rhynchobdellida			
Glossiphoniidae	4	1	
Pharyngobdellida			
Erpobdellidae	2		
Mollusca			
Gastropoda			
Basommatophora			
Hydrobiidae			
Lymnaeidae	2	1	1
Planorbidae	33	3	
Physidae	53	4	1
Pelecypoda			
Heterodonta			
Pisidiidae	3		
Arthropoda			
Crustacea			
Amphipoda			
Talitridae		1	3
Insecta			
Coleoptera			
Dytiscidae	4		
Haliplidae	6		
Helodidae		4	
Hydrophilidae	23		
Diptera			
Chironomidae			
Chironominae	1	1	13
Tanypodinae			1
Ceratopogonidae			1
Culicidae (Chaoborinae)			
Empidae		1	
Stratiomyidae			
Ephemeroptera			
Baetidae	17		2
Caenidae	7		8
Heptageniidae	2		
Hemiptera			
Belostomatidae			
Corixidae	8		
Lepidoptera			
Pyrallidae			
Odonata (Anisoptera)			
Corduliidae			
Libellulidae			
Odonata (Zygoptera)			
Coenagrionidae			1

**Appendix D**

**Site Water Quality Data**

Table 13. Site water quality data collected from the Yellowbank Slough region during field sampling in 1991.

Sample Location	Sample Number	Temp (C°)	pH	DO (ppm)	Specific Conductance (umhos)	Salinity (%)
Upper Yellowbank Slough (upstream reference site)	YS-1	32.0	7.3	4.0	450.0	0.0
Upper Yellowbank Slough	YS-2	No data collected.				
Middle Yellowbank Slough	YS-3	26.0	7.4	2.8	600.0	0.0
Lower Yellowbank Slough	YS-4	27.0	8.1	>15.0	750.0	0.0
Lower Yellowbank Slough	YS-5	28.0	8.0	>15.0	675.0	0.0
Lower Running Slough	YS-6	28.0	7.9	13.2	650.0	0.0
Middle Running Slough	YS-7	31.0	7.9	1.1	600.0	0.0
Middle Running Slough	YS-8	No data collected.				
Goose Pond	YS-9	33.5	7.5	1.2	370.0	0.0
Lower Running Slough	YS-10	No data collected.				
Crude Oil Spill Site (positive control)	YS-11	No data collected.				
Shawneetown Vicinity (negative control)	YS-12	26.8	7.2	6.1	445.0	0.0

## **Appendix E**

### **Toxicity Test Data: Aquatic Invertebrates**

Table 14(a). 1991 Yellowbank Slough 96-hour Ceriodaphnia dubia bioassay results  
(total mortality after 96 hours of exposure).

Sample Location	Sample Number	<u>Ceriodaphnia dubia</u> mortality (number dead/number tested):	
		Control	Sample
Upper Yellowbank Slough (reference site)	YS-1	0/20	0/20
Lower Yellowbank Slough	YS-4	0/20	0/20
Middle Running Slough	YS-7	0/20	0/20
Goose Pond	YS-9	0/20	0/20
Shawneetown Vicinity (reference site)	YS-12	0/20	0/21



Table 14(b). Water chemistry data for 96-hour Ceriodaphnia dubia bioassays using surface water collected from the Yellowbank Slough region in 1991.

Sample Location	Sample Number	Temp (C°)	pH	DO (ppm)	NH <sub>3</sub> -N (mg/L)	Total NH <sub>3</sub> (mg/L)	Unionized NH <sub>3</sub> (mg/L)	Total Hardness (mg/L CaCO <sub>3</sub> )	Specific Conductance (umhos)	Total Alkalinity	Total Residual Chlorine
Upper Yellowbank Slough (upstream reference site)	YS-1	25.0	8.0	8.8	0.1	<0.12	<0.01	239.0	220.0	217.0	<0.005
Lower Yellowbank Slough	YS-4	25.0	8.2	8.6	<0.1	<0.12	<0.01	342.0	840.0	278.0	<0.005
Middle Running Slough	YS-7	25.0	7.8	7.7	<0.1	<0.12	<0.01	247.0	490.0	224.0	<0.005
Goose Pond	YS-9	25.0	7.8	6.9	<0.1	<0.12	<0.01	225.0	440.0	201.0	<0.005
Shawneetown Vicinity (reference site)	YS-12	25.0	8.1	7.2	<0.1	<0.12	<0.01	238.0	500.0	203.0	<0.005

Table 15(a). 1991 Yellowbank Slough solid phase sediment and water 10-day Myallela azteca bioassay results (total mortality after 10 days of exposure).

Sample Location	Sample Number	<u>Myallela azteca</u> mortality (number dead/number tested):			Total Mortality (%)
		Test Replicate			
		<u>A</u>	<u>B</u>	<u>C</u>	
Upper Yellowbank Slough (upstream reference site)	YS-1	0/20	0/20	0/20	0/60 (0)
Lower Yellowbank Slough	YS-4	1/20	5/20 <sup>1</sup>	1/20	2/40 (5)
Middle Running Slough	YS-7	0/20	0/20	0/20	0/60 (0)
Goose Pond	YS-9	0/20	0/20	0/20	0/60 (0)
Shawneetown Vicinity (reference site)	YS-12	2/20	1/10	0/20	3/60 (5)
[ Laboratory Control		0/20	0/20	0/20	0/60 (0) ]

<sup>1</sup>Replicate excluded from analysis due to insufficient recovery of organisms.

Table 15(b). Water chemistry data for solid-phase sediment and water 10-day Hyallela azteca bioassays using sediments collected from the Yellowbank Slough region in 1991.

Sample Location	Sample Number	Temp (C°)	pH	DO (ppm)	NH <sub>3</sub> -N (mg/L)	Total NH <sub>3</sub> (mg/L)	Unionized NH <sub>3</sub> (mg/L)	Total Hardness (mg/L CaCO <sub>3</sub> )	Specific Conductance (umhos)	Total Alkalinity	Total Residual Chlorine
Upper Yellowbank Slough	YS-1	25.0	8.0	8.8	0.1	<0.12	<0.01	239.0	220.0	217.0	<0.005
Lower Yellowbank Slough	YS-4	25.0	8.2	8.6	<0.1	<0.12	<0.01	342.0	840.0	278.0	<0.005
Middle Running Slough	YS-7	25.0	7.8	7.7	<0.1	<0.12	<0.01	247.0	490.0	224.0	<0.005
Goose Pond	YS-9	25.0	7.8	6.9	<0.1	<0.12	<0.01	225.0	440.0	201.0	<0.005
Shawneetown Vicinity (reference site)	YS-12	25.0	8.1	7.2	<0.1	<0.12	<0.01	238.0	500.0	203.0	<0.005

## Appendix F

### Toxicity Test Data: Microtox™

Table 16. 1991 Yellowbank Slough Microtox toxicity test results.

Sample Location	Sample Number	Solid-Phase	100%
		Test EC 50	Assay Test EC 50
Upper Yellowbank Slough (upstream reference site)	YS-1	0.2090	No toxicity detected
Middle Yellowbank Slough	YS-3	0.0789	No toxicity detected
Lower Yellowbank Slough	YS-4	0.6069	No toxicity detected
Lower Yellowbank Slough	YS-5	0.2662	No toxicity detected
Lower Running Slough	YS-6	0.1977	No toxicity detected
Middle Running Slough	YS-7	0.1021	No toxicity detected
Goose Pond	YS-9	0.1021	No toxicity detected
Shawneetown Vicinity (reference site)	YS-12	2.1046	No toxicity detected

# MICROTOX DATA REPORT

FILE NAME: B:\YS1B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

Sample Description:

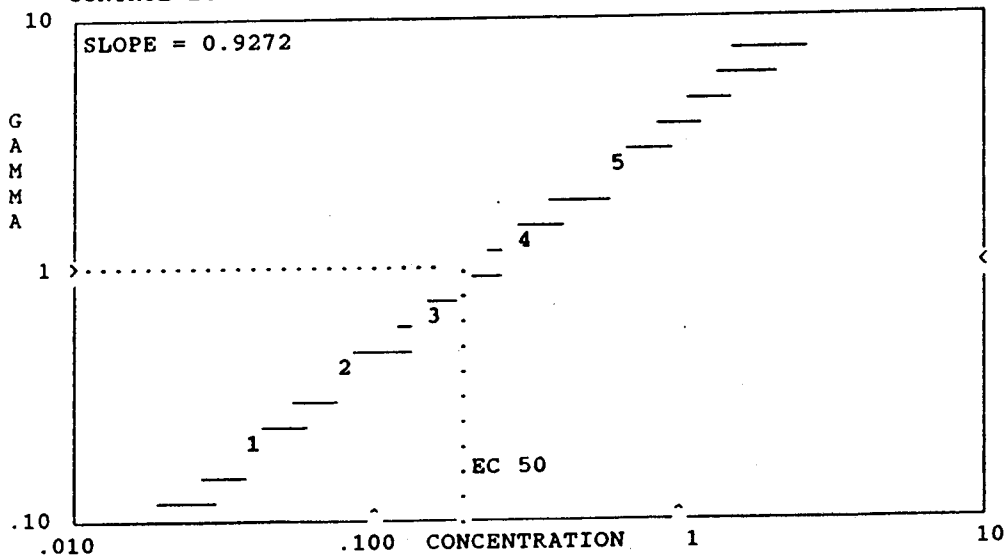
Yellowbank Slough bottom sediment sample #YS-1, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	80	0.0391	0.21250#
2	69	0.0781	0.40580#
3	55	0.1563	0.76364#
4	41	0.3125	1.36585#
5	25	0.6250	2.88000#
6	11	1.2500	7.81818
7	3	2.5000	31.33333
8	1	5.0000	96.00000
9	1	10.0000	96.00000

CONTROL It's : 97



EC 50 = 0.2090 (95% CONFIDENCE RANGE: 0.1945 TO 0.2246)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS3B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough bottom sediment sample #YS-3, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE

Initial Concentration : 10 %

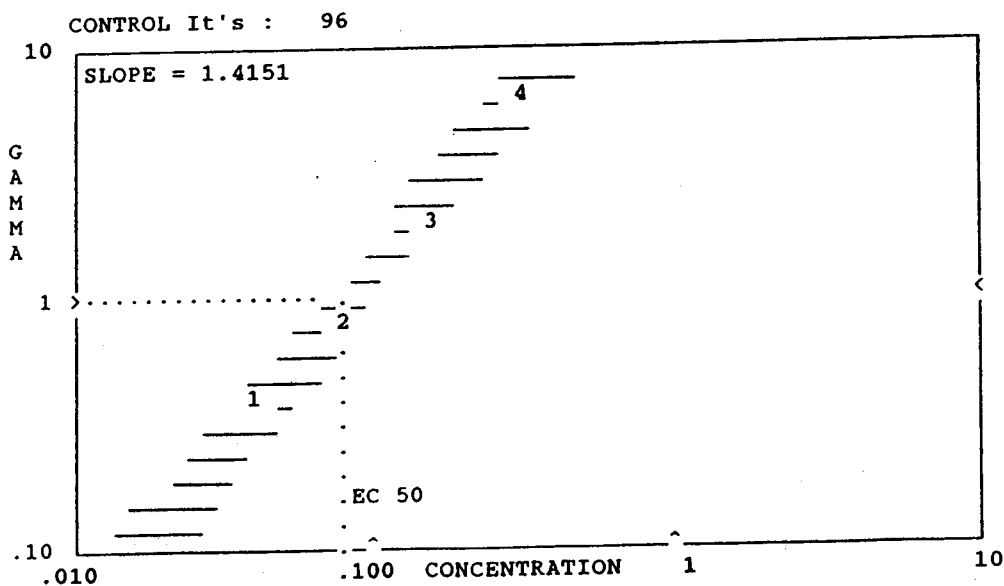
Assay Time: 20 minutes

Ionic Adjustment: NO

Dilution Factor : 2

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	68	0.0391	0.41176#
2	51	0.0781	0.88235#
3	28	0.1563	2.42857#
4	11	0.3125	7.72727#
5	0	0.6250	> 999 *
6	0	1.2500	> 999 *
7	0	2.5000	> 999 *
8	0	5.0000	> 999 *
9	0	10.0000	> 999 *



EC 50 = 0.0789 (95% CONFIDENCE RANGE: 0.0626 TO 0.0996)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS4B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

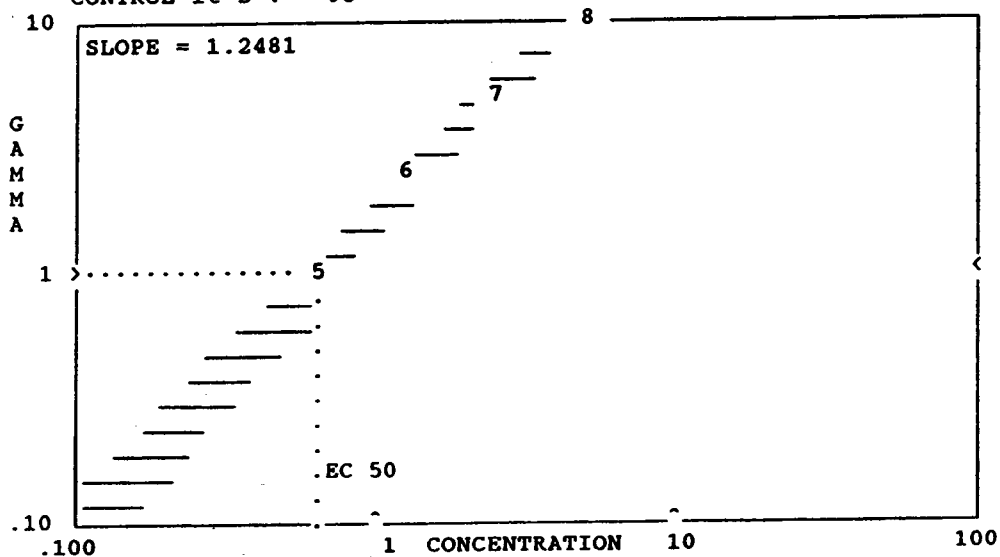
Yelloebank Slough bottom sediment sample #YS-4, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	86	0.0391	0.10465
2	81	0.0781	0.17284
3	58	0.1563	0.63793
4	52	0.3125	0.82692
5	46	0.6250	1.06522#
6	27	1.2500	2.51852#
7	15	2.5000	5.33333#
8	6	5.0000	14.83333#
9	3	10.0000	30.66667

CONTROL It's : 95



EC 50 = 0.6069 (95% CONFIDENCE RANGE: 0.4750 TO 0.7753)

# Used for calculations



# MICROTOX DATA REPORT

FILE NAME: B:\YS5B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

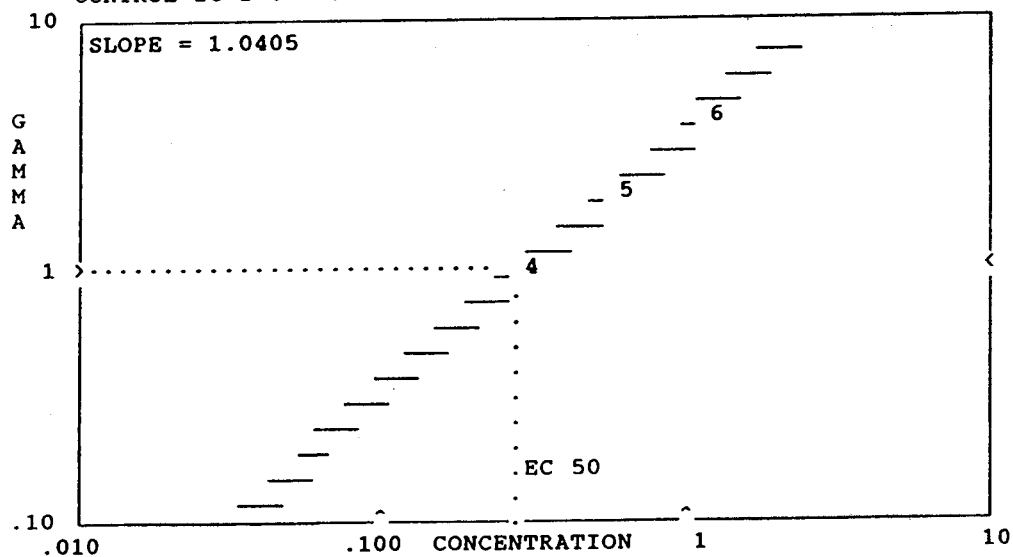
Yellowbank Slough bottom sediment sample #YS-5, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	65	0.0391	0.47692
2	66	0.0781	0.45455
3	57	0.1563	0.68421
4	44	0.3125	1.18182#
5	28	0.6250	2.42857#
6	16	1.2500	5.00000#
7	6	2.5000	15.00000
8	4	5.0000	23.00000
9	3	10.0000	31.00000

CONTROL It's : 96



EC 50 = 0.2662 (95% CONFIDENCE RANGE: 0.2647 TO 0.2678)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS6B8591.SPT

TEST DATE: \_\_\_\_\_  
TEST TIME: \_\_\_\_\_

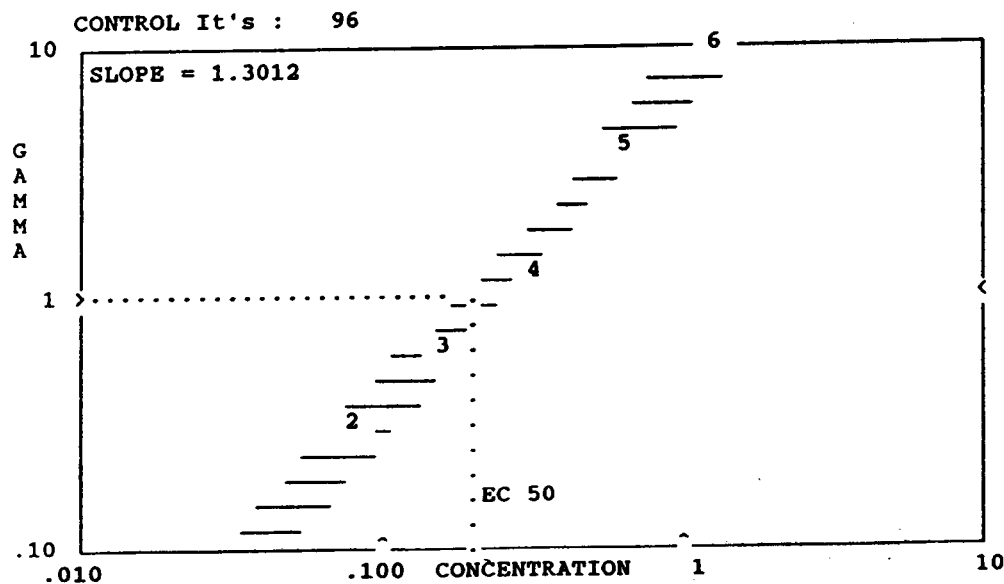
## Sample Description:

Yellowbank Slough bottom sediment sample #YS-6, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	72	0.0391	0.33333
2	71	0.0781	0.35211#
3	57	0.1563	0.68421#
4	38	0.3125	1.52632#
5	18	0.6250	4.33333#
6	7	1.2500	12.71429#
7	1	2.5000	95.00000
8	0	5.0000	> 999 *
9	0	10.0000	> 999 *



EC 50 = 0.1977 (95% CONFIDENCE RANGE: 0.1618 TO 0.2416)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS7B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

Sample Description:

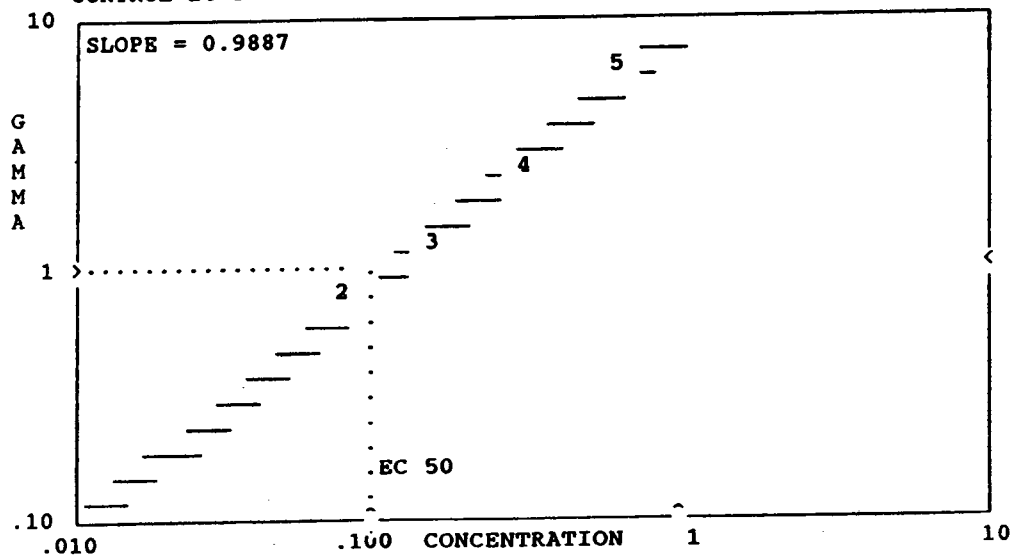
Yellowbank Slough bottom sediment sample YS-7, subsample "b"  
, tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	62	0.0391	0.54839
2	53	0.0781	0.81132#
3	39	0.1563	1.46154#
4	25	0.3125	2.84000#
5	13	0.6250	6.38462#
6	5	1.2500	18.20000
7	1	2.5000	95.00000
8	1	5.0000	95.00000
9	1	10.0000	95.00000

CONTROL It's : 96



EC 50 = 0.1021 (95% CONFIDENCE RANGE: 0.0859 TO 0.1215)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS9B8591.SPT

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

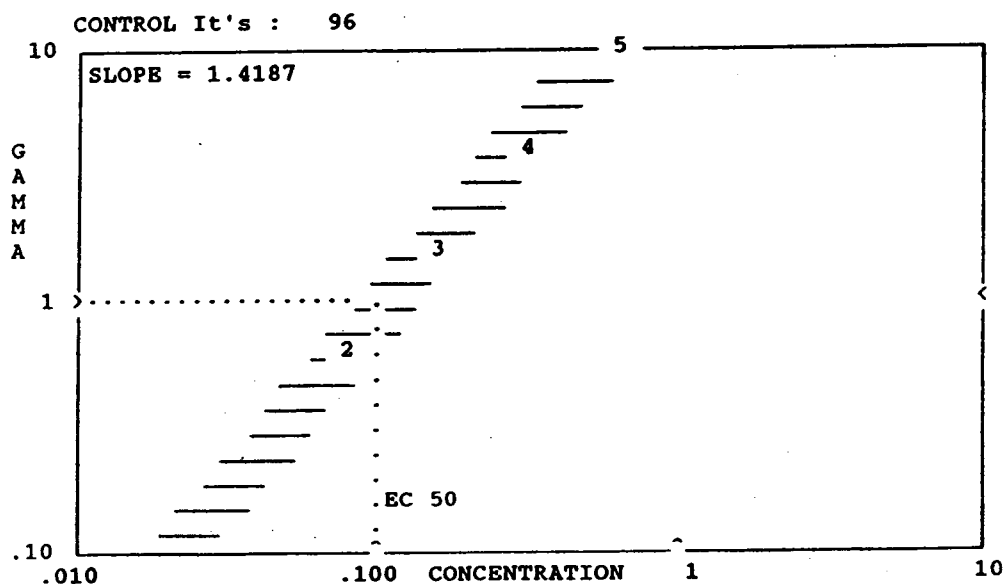
Sample Description:

Yellowbank Slough bottom sediment sample YS-9, subsample "b"  
, tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	60	0.0391	0.60000
2	54	0.0781	0.77778#
3	36	0.1563	1.66667#
4	18	0.3125	4.33333#
5	6	0.6250	15.00000#
6	1	1.2500	95.00000
7	0	2.5000	> 999 *
8	0	5.0000	> 999 *
9	0	10.0000	> 999 *



EC 50 = 0.1021 (95% CONFIDENCE RANGE: 0.0789 TO 0.1321)

# Used for calculations

# MICROTOX DATA REPORT

FILE NAME: B:\YS12B859.SPT

TEST DATE: \_\_\_\_\_  
TEST TIME: \_\_\_\_\_

## Sample Description:

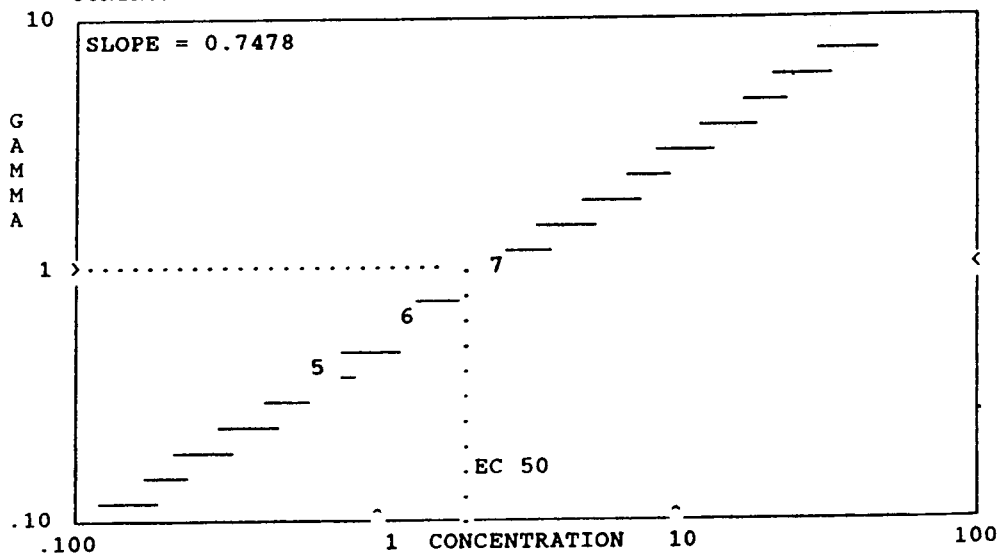
Yellowbank Slough bottom sediment sample YS-12, subsample "b", tested 8/5/91.

Procedure: SOLID-PHASE  
Initial Concentration : 10 %  
Assay Time: 20 minutes

Ionic Adjustment: NO  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	95	0.0391	-0.01053*
2	88	0.0781	0.06818
3	79	0.1563	0.18987
4	82	0.3125	0.14634
5	67	0.6250	0.40299#
6	56	1.2500	0.67857#
7	44	2.5000	1.13636#
8	24	5.0000	2.91667
9	11	10.0000	7.54545

CONTROL It's : .94



EC 50 = 2.1046 (95% CONFIDENCE RANGE: 2.0424 TO 2.1686)

# Used for calculations  
\* Invalid gammas

# MICROTOX DATA REPORT

FILE NAME: B:\YS3A8791.E20

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough pore water sample extracted from bottom sediment sample #YS-3, subsample "a", tested 8/7/91.

Procedure: 100%

Initial Concentration : 91 %

Assay Time: 20 minutes

Ionic Adjustment: MOAS

Dilution Factor : 2

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	89	11.3750	-0.14607*
2	90	22.7500	-0.15556*
3	89	45.5000	-0.14607*
4	89	91.0000	-0.14607*

CONTROL It's : 76

EC 50 IS GREATER THAN 100%

\* Invalid gammas

# MICROTOX DATA REPORT

FILE NAME: B:\YS4A8791.E20

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough pore water extracted from bottom sediment  
sample #YS-4, subsample "a", tested 8/7/91.

Procedure: 100%

Initial Concentration : 91 %

Assay Time: 20 minutes

Ionic Adjustment: MOAS

Dilution Factor : 2

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	86	11.3750	-0.06977*
2	85	22.7500	-0.05882*
3	78	45.5000	0.02564*
4	60	91.0000	0.33333#

CONTROL It's : 80

EC 50 IS GREATER THAN 100%

\* Invalid gammas

# MICROTOX DATA REPORT

FILE NAME: B:\YS5A8791.E20

TEST DATE: \_\_\_\_\_

Sample Description:

TEST TIME: \_\_\_\_\_

Yellowbank Slough pore water sample extraction from bottom sediment sample #YS-5, subsample "a", tested 8/7/91.

Procedure: 100%

Ionic Adjustment: MOAS

Initial Concentration : 91 %

Dilution Factor : 2

Assay Time: 20 minutes

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	74	11.3750	0.20270#
2	71	22.7500	0.25352#
3	58	45.5000	0.53448#
4	64	91.0000	0.39063#

CONTROL It's : 89

EC 50 IS GREATER THAN 100%  
IS GREATER THAN 100%

# Used for calculations



# MICROTOX DATA REPORT

FILE NAME: B:\YS6A8791.E20

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough pore water sample extraction from bottom sediment sample #YS-6, subsample "a", tested 8/7/91.

Procedure: 100%

Initial Concentration : 91 %

Assay Time: 20 minutes

Ionic Adjustment: MOAS

Dilution Factor : 2

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	83	11.3750	-0.09639*
2	86	22.7500	-0.12791*
3	78	45.5000	-0.03846*
4	45	91.0000	0.66667#

CONTROL It's : 75

EC 50 IS GREATER THAN 100%

\* Invalid gammas

# MICROTOX DATA REPORT

FILE NAME: B:\YS7B8791.E20

TEST DATE: \_\_\_\_\_  
TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough pore water sample extracted from bottom sediment sample #YS-7, subsample "b" (subsample "a" not tested due to insufficient pore water), tested 8/7/91.

Procedure: 100%  
Initial Concentration : 91 %  
Assay Time: 20 minutes

Ionic Adjustment: MOAS  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	87	11.3750	-0.06897*
2	75	22.7500	0.08000#
3	73	45.5000	0.10959#
4	64	91.0000	0.26563#

CONTROL It's : 81  
EC 50 IS GREATER THAN 100%  
IS GREATER THAN 100%

# Used for calculations  
\* Invalid gammas

# MICROTOX DATA REPORT

FILE NAME: B:\YS9A8791.E20

TEST DATE: \_\_\_\_\_  
TEST TIME: \_\_\_\_\_

## Sample Description:

Yellowbank Slough pore water sample extraction from bottom sediment sample #YS-9, subsample "a", tested 8/7/91.

Procedure: 100%  
Initial Concentration : 91 %  
Assay Time: 20 minutes

Ionic Adjustment: MOAS  
Dilution Factor : 2  
Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	88	11.3750	-0.02273*
2	78	22.7500	0.10256#
3	66	45.5000	0.30303#
4	58	91.0000	0.48276#

CONTROL It's : 86

EC 50 IS GREATER THAN 100%  
IS GREATER THAN 100%

# Used for calculations  
\* Invalid gammas

MICROTOX DATA REPORT

FILE NAME: B:\YS12A879.E20

TEST DATE: \_\_\_\_\_

TEST TIME: \_\_\_\_\_

Sample Description:

Yellowbank Slough pore water sample extraction from sample #  
YS-12, subsample "a", tested 8/7/91.

Procedure: 100%

Initial Concentration : 91 %

Assay Time: 20 minutes

Ionic Adjustment: MOAS

Dilution Factor : 2

Concentration Units: %

NUMBER	It	CONC.	GAMMA
1	116	11.3750	-0.18103*
2	103	22.7500	-0.07767*
3	94	45.5000	0.01064*
4	91	91.0000	0.04396*

CONTROL It's : 95

EC 50 IS GREATER THAN 100%

\* Invalid gammas